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PROCEEDINGS OF THE
SECOND INTERNATIONAL WORKSHOP
ON ALLIUM WHITE ROT

JUNE 22-24, 1983 / #c

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Sponsored by
The Soilborne Diseases Laboratory,
Plant Protection Institute,
Agricultural Research Center,
U.S. Department of Agriculture
Beltsville, Maryland 20705

Workshop Coordinator: Peter B. Adams

Program Chairman: Andrew R. Entwistle

SECOND INTERNATIONAL WORKSHOP
ON ALLIUM WHITE ROT
PROGRAM

Wednesday, June 22, 1983

8:00-8:30 AM Registration: P. B. Adams

8:30-9:00 AM I. General Welcome: G. C. Papavizas
P. A. Putnam, Director, BARC

9:00-9:30 AM II. Progress of AWRG and Information Letter
1979-1983, A. R. Entwistle and P. B. Adams

9:30-12:00 AM III. Allium White Rot: Geographic Distribution
and Economic Importance

Chairman: P. B. Adams

12:00-1:30 PM Lunch - Local restaurants or USDA Cafeteria

1:30-4:30 PM IV. Chemical Control

Chairman: A. R. Entwistle

Introduction: "Needs and outlook for chemical
control," Entwistle.

Practical results 1979-1983, Mode of action of
control measures.

Fungicide tolerance in Sclerotium cepivorum.
Contribution by participants.

5:00-7:30 PM Dinner - Local restaurants

7:30-9:30 PM V. Demonstration of Methods

Chairman: P. B. Adams

MEMORANDUM FOR THE RECORD

TO: THE BOARD OF TRUSTEES

DATE: 1950-11-15

FROM: THE BOARD OF TRUSTEES

DATE: 1950-11-15

SUBJECT: THE BOARD OF TRUSTEES

1. The Board of Trustees of the University of Chicago has the honor to acknowledge the receipt of the letterhead memorandum from the Board of Trustees of the University of Illinois, dated November 10, 1950, regarding the proposed merger of the two universities.

2. The Board of Trustees of the University of Chicago has considered the proposed merger and has concluded that it is in the best interests of the University to accept the offer of merger.

3. The Board of Trustees of the University of Chicago has authorized the President of the University to execute the necessary documents to effect the merger.

4. The Board of Trustees of the University of Chicago has authorized the President of the University to execute the necessary documents to effect the merger.

5. The Board of Trustees of the University of Chicago has authorized the President of the University to execute the necessary documents to effect the merger.

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12. The Board of Trustees of the University of Chicago has authorized the President of the University to execute the necessary documents to effect the merger.

Thursday, June 23, 1983

8:00-12:00 Noon VI. Genetic Control

Chairman: Q. P. van der Meer

12:00-1:30 PM Lunch, local restaurants or USDA Cafeteria

1:30-4:30 PM VII. Biological Control

Chairman: J. E. Rahe

5:00-7:30 PM Dinner - Local restaurants

7:30-9:30 PM VIII. Epidemiology and Disease Forecasting

Chairman: D. H. Hall

Friday, June 24, 1983

8:00-11:00 AM IX. Integrated Control

Chairman: S. A. Johnston

Introduction: "State of the Science-future
needs and future trends," Coley-Smith.

11:00-12:00 Noon X. Closing Discussion

Chairman: G. C. Papavizas

Future research

Third Workshop: 1986

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A.R. Entwistle

The objectives of the First Workshop were to:-

- Optimise communications between those interested in onion white rot.
- Prepare a world list of scientists active and interested in onion white rot.
- Publish a Newsletter
- Organise Workshops probably at three-year intervals.
- Compile an international white rot differential for use in resistance screening breeding experiments.
- Prepare a bibliography on onion white rot.

It is appropriate that we should review progress so that members may decide on any improvements. Below I list my impressions on some aspects.

1. List of white rot workers. The first list included all names sent to me. A revised list has now been produced which includes only those who replied to a Questionnaire sent with Newsletter No. 2. The intention is to restrict the list to 'active' workers and those who have an active interest.

2. Information letters (formerly newsletters). The first two issues were published by A.R. Entwistle in December 1980 and March 1982. Dr. Adams then became Editor and published Number 3 in October 1982. Of the four suggestions made in Newsletter 1, viz.

- short summaries of recent work and ideas.
- details of personal and academic achievements and of visits etc.
- thoughts about the next Workshop
- an article on resistance screening,

research summaries and workshop details have formed the main contents.

3. Workshops General opinion was that 1983 rather than 1982 was best for the 2nd workshop. Several correspondents expressed the desire that attendance be kept reasonably small to favour a lively atmosphere.

The venue and date of the Third workshop are to be decided in Session X.

4. White rot differential. Members felt that it was too early to compile a list (Newsletter 2).

5. Bibliography. The first computerized bibliography was produced by A.R. Entwistle in autumn 1980 and about 30 copies were sent on request. Several requests have come from Libraries in addition to white rot workers. New references are listed in a separate file for periodic updating of the main file.

There has been delay in the last update while NVRS (UK) computing facilities are restructured.

It is very helpful when authors send details of their publications as soon as they have been accepted, since page details etc. can be revised at a later date.

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Allium White Rot in the United States

P. B. Adams

Plant Pathologist, Soilborne Diseases Laboratory, Plant Protection
Institute, U.S. Department of Agriculture, Beltsville, MD 20705

White rot caused by Sclerotium cepivorum Berk. was first observed in the United States in Oregon in 1918 and later in Virginia in 1923 (5). It has now been reported in California, Illinois, Kentucky, Louisiana, New Jersey, Oregon, Virginia, and Washington (2). Within the past two years the disease has been found in Nevada and more recently has been observed in New York. The disease appears to be of economic importance only in California, Nevada, New Jersey, Oregon, and Washington.

In Lyon County, Nevada, bulb onions (A. cepa L.) are grown for the fresh market and garlic (A. sativum L.) is grown for seed production. A large portion of the nation's garlic seed is produced in this county. The two crops are grown on 525-610 ha and is estimated to be worth 5-6 million dollars. White rot is causing severe losses on these crops, and without effective control measures, it will increase.

In New Jersey bulb onions are grown on 320 ha with a value of approximately \$1.4 million (4). This crop suffers very little loss to white rot because it is grown during the summer when the soil temperatures are too high for infection by S. cepivorum. Leek (A. porrum L.) and bunching onions (scallions) (A. cepa and A. cepa X A. fistulosum amphidiploid) are grown on 200 ha with a market value of \$4.3 million. Most of these crops are grown in Atlantic, Camden, Cumberland, Gloucester and Salem counties. The crops are either directly seeded or transplanted in the fall and harvested in the spring. Under these conditions

white rot causes severe losses due to thinning of the plant stand in the fall months and diseased plants in the spring months (3). The amount of white rot on these two crops at harvest range from 0-100% but average about 20% or a \$1 million loss. This loss does not take into account the loss due to the reduced plant stand that occurs during the fall months. Until recently the only economical control measure available to the growers was crop rotation which most farmers practiced. Iprodione is now used by some farmers while others fumigate their fields with metham applied through sprinkler irrigation before planting.

In Orange County, New York white rot has been a "problem" on early spring-planted bulb onions on one farm. This farmer does not practice crop rotation but has fumigated his fields every year for the past 9 years (S. E. Todd, personal communication).

Oregon bulb onions valued at \$48.2 million are grown on 3800 ha (4). White rot is present in a small area in Marion County in the western part of the state and in Umatilla County in the northeastern part of the state. Losses due to white rot are not economically severe at this time but could become so in the near future.

Washington grows bulb onions on 1880 ha and the crop is valued at \$6.3 million. White rot appears to be restricted to about 320 ha in Walla Walla County. The onions grown in this area are a special variety with a very mild flavor and thus the farmers are paid a premium price (\$3.5 million) for the commodity. White rot is estimated to cause approximately 50% loss each year or about \$1.8 million. In addition, 120 ha of good land has been lost to onion production due to white rot. Farmers are now moving their onion production to new, less fertile land. They have found that this new land will economically produce onions for about 5 years before the infestation of S. cepivorum becomes too severe (V. Criscola, personal communication).

1 A conservative estimate of the losses of Allium commodities in New Jersey
2 and Washington alone due to white rot is \$2.8 million per year. Losses in
3 Nevada are unknown and in Oregon appear to be very small; however, in both
4 States they could become severe in the near future.

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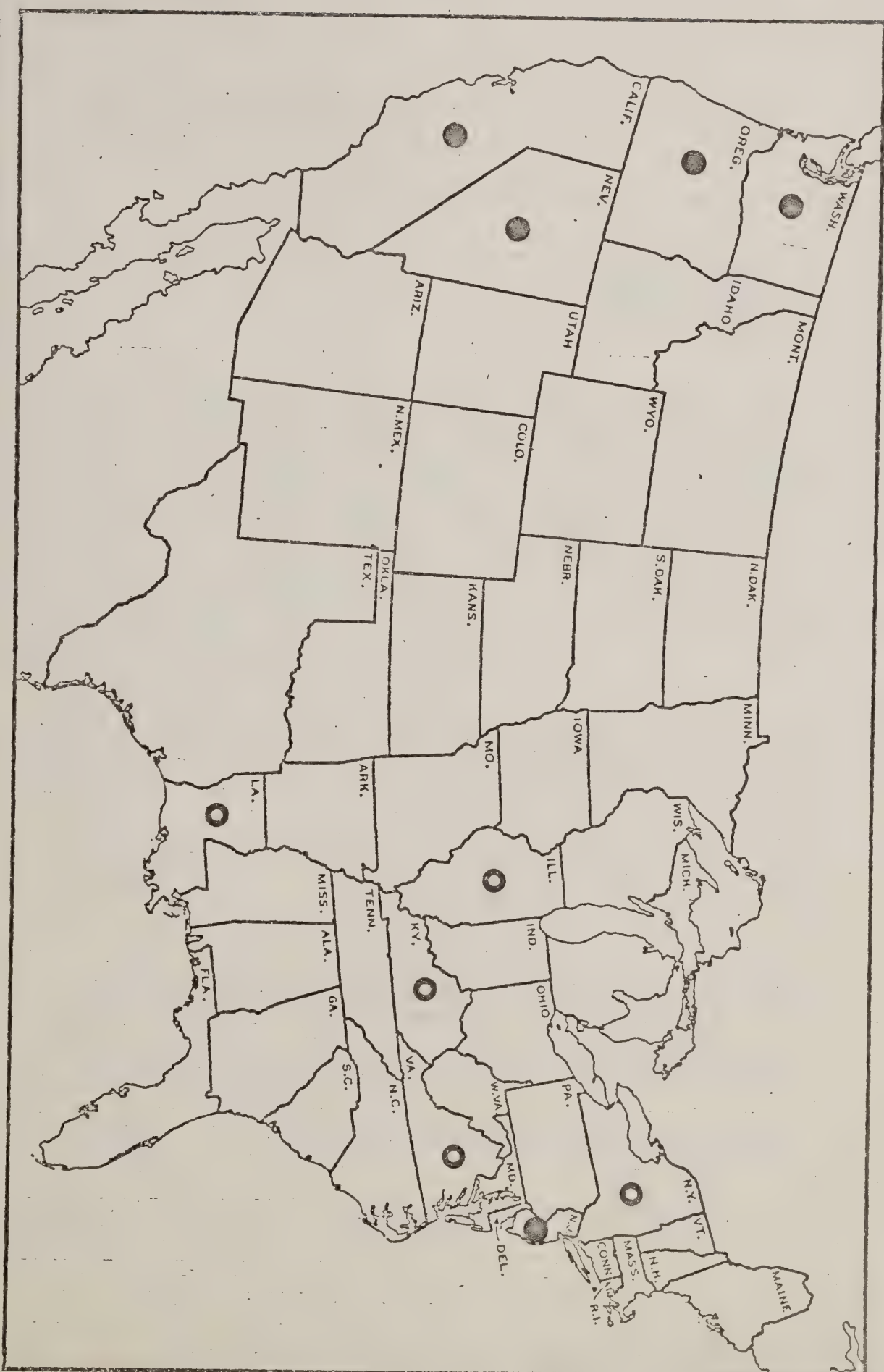


Figure 1. Geographic distribution of Allium White Rot in the United States. Open circles represent states in which white rot is known to occur. Solid circles represent states in which white rot causes economic losses to crops.

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Second International Workshop
on Allium White Rot
Agricultural Research Center
Beltsville, Maryland, U.S.A.

Session III. Allium White Rot: Geographic
Distribution and Economic
Importance

Allium White Rot In USA

S. A. Johnston
Rutgers University
Bridgeton, N.J.

In New Jersey, there are several onion types grown. These include the dry bulb onions, 265 ha; bunching onions, 100 ha; leeks, 60 ha; and garlic. The dry bulb onions are planted (seeds or sets) during March and April and harvested from June to October. Bunching onions are seeded (11 kg seed/ha) in rows that are 0.3 meters apart during March and April for summer harvest or they are seeded in August and early September for late fall harvest or overwintered and harvested during the spring. Leeks produced from seed in the Southern U.S. are transplanted in early spring for summer harvests, or they are produced from seed in seedbeds in N.J. and transplanted into production fields in rows spaced 0.6 meters apart and 0.15 meters between plants for late fall harvest or overwintered for spring harvest.

White Rot incidence is only experienced in the overwintered crops of bunching onions and leeks. Losses are estimated at 5% of the overwintered onions and leeks crop with losses of 80-90% experienced in some fields. Disease incidence is much higher in years with cool, moist springs.

Research efforts during the past four years have been in the following areas:

1. Chemical Control
 - a. Fungicide evaluations involving soil applications in the fall with foliar applications in the spring.
 - b. The application of metham through irrigation systems in summer prior to late summer seedings.
 - c. Addition of fungicides to pregerminated seed in fluid drilled seedings.
2. Biological control - evaluation of fungal antagonists applied at seeding.

Economic Importance and Distribution of 'White Rot' of Onions and Garlic in California

Prudence A. Somerville and Dennis H. Hall

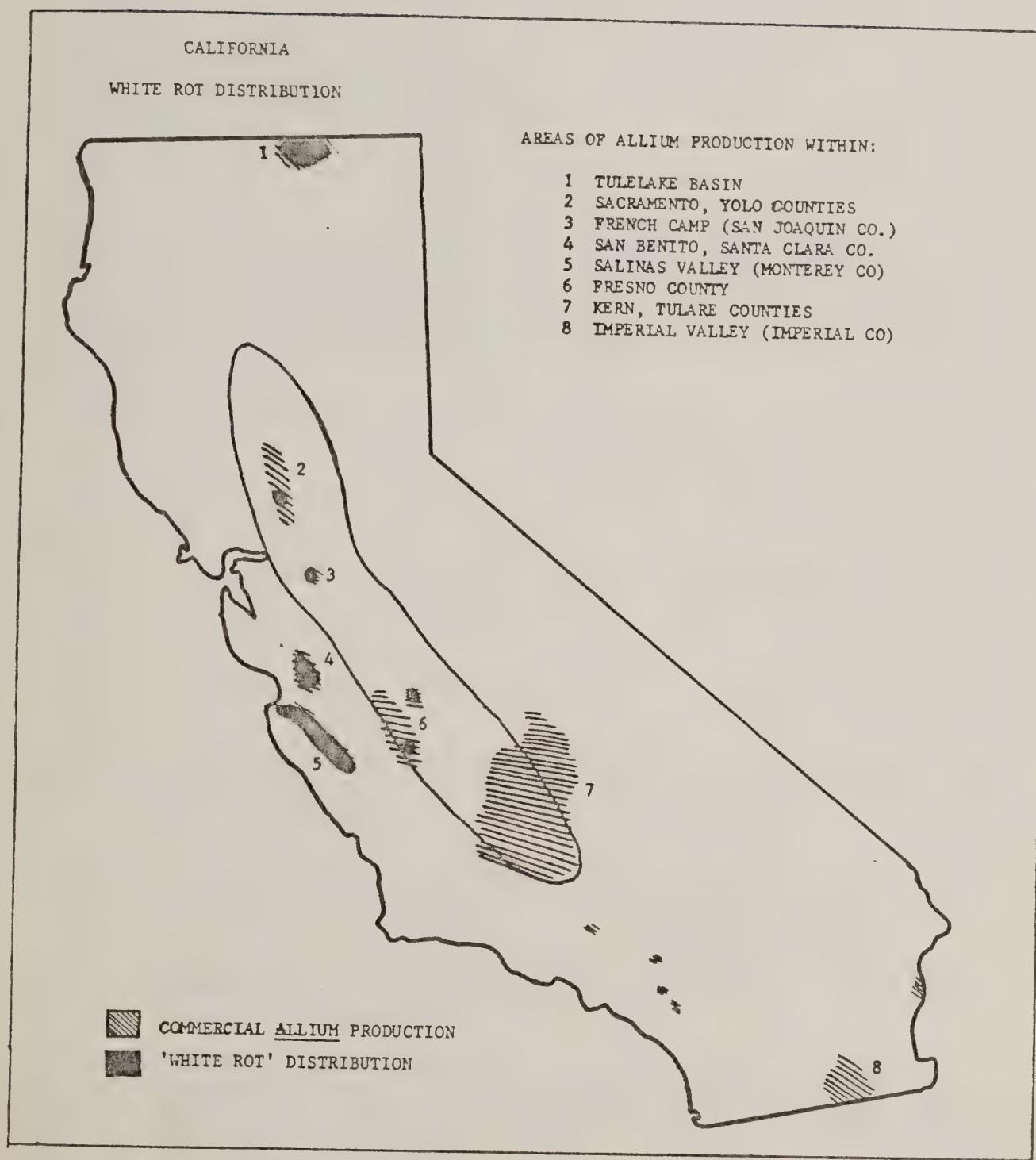
Department of Plant Pathology, University of California, Davis, CA

White rot was first reported in California in the central coast counties of San Mateo and San Benito. The disease has now been found in all of the major Allium production areas, except Kern County and the desert area, Imperial Valley.

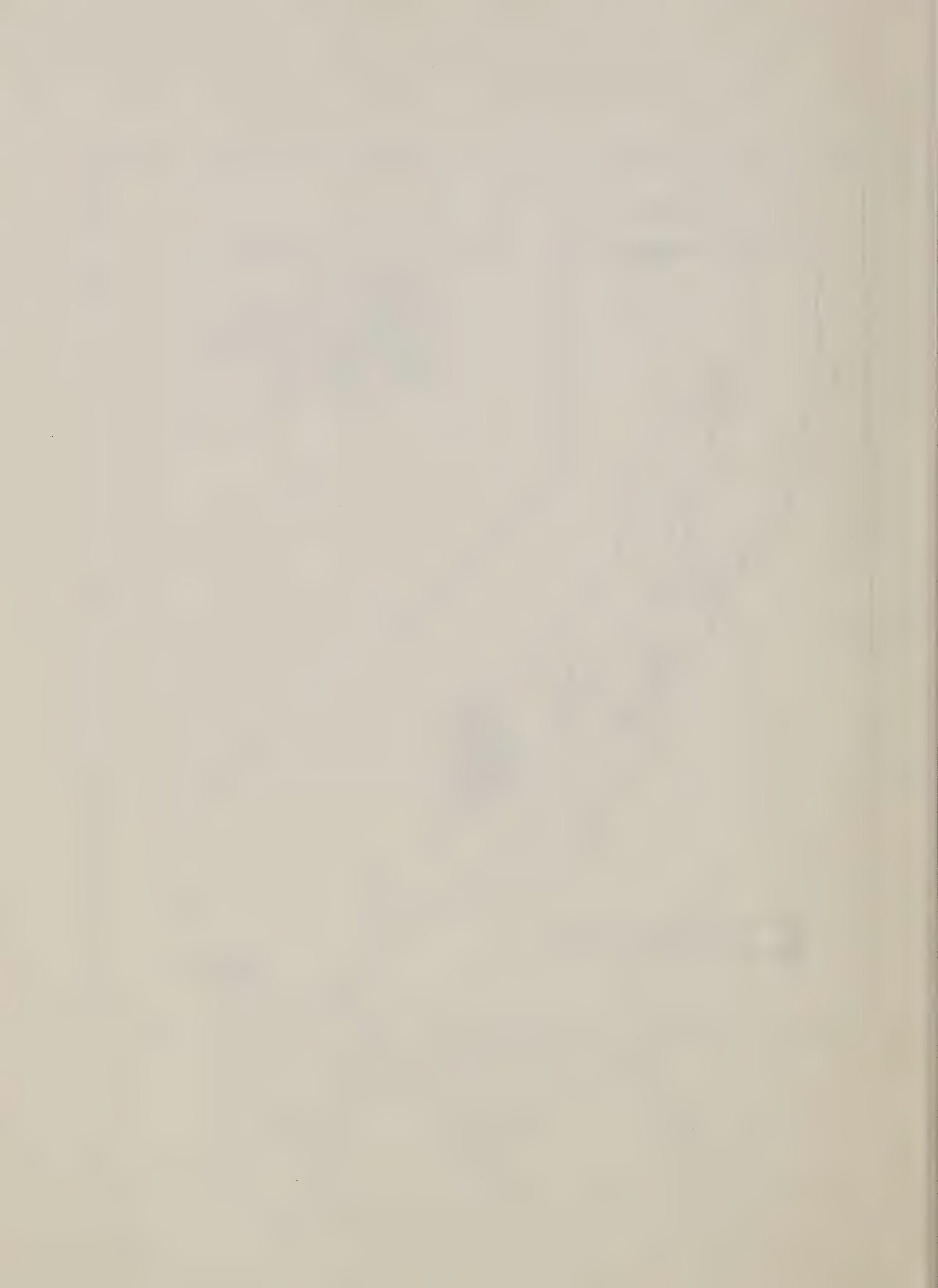
White rot was reported in the Tularelake Basin in 1957 and has been found in many fields since then. In San Benito and Monterey counties 'white rot' is widespread and of economic importance. Growers avoid infested fields. In Fresno County, isolated outbreaks have been reported since 1976. The spread of 'white rot' within San Joaquin County was due to the sale of infected onion transplants.

At Davis (Yolo County) 'white rot' was introduced when soil was artificially inoculated for experimental use.





1981 California <u>Allium</u> Statistics		
<u>Crop</u>	<u>Area (ac)</u>	<u>Value (\$M)</u>
Garlic	11,050	27.3
Onion	33,200	128.8



SECOND INTERNATIONAL WORKSHOP ON ALLIUM WHITE ROT

ABSTRACT - GEOGRAPHIC DISTRIBUTION AND ECONOMIC IMPORTANCE

EDWARD A. KURTZ
TECHNICAL DIRECTOR
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BASIC VEGETABLE PRODUCTS DIVISION
700 AIRPORT DRIVE
KING CITY, CALIFORNIA
93930

White rot (*Sclerotium cepivorum*) is a serious disease and can create economic losses to garlic and onions in certain areas of California. The two primary areas currently infested with this disease are the Central Coast (Monterey, San Benito and Santa Clara Counties) and the northern area (Modoc and Siskiyou Counties).

The following data is presented for California:

<u>CROP</u>	<u>AREA</u>	<u>ACRES/VALUE</u>	
		<u>1981</u>	<u>1982</u>
Garlic	Central	2,330/\$12,682,000	2,604/\$9,303,000
Onion	Central	2,540/\$10,430,000	2,340/\$4,467,000
Onion	Northern	975/\$1,630,300	1,912/\$3,569,900

Approximately 25-30% of the garlic acreage in the Central Coast area was treated with Botran as and in-furrow spray at planting in the fall of 1982 and again in 1983.

Although the total loss of individual fields has been recorded, a more common occurrence is for portions of fields to be lost due to "hot spots" arising from high populations of *S. cepivorum* sclerotia.

The following formula can be used to determine individual field losses resulting from white rot infestations:

Average number of plants per bed foot times estimated bed feet lost times estimated bulb weight (garlic 40 grams/bulb - Onions 65 grams/bulb) ÷ 454 equals estimated lost pounds/acre times value/pound equals estimated dollar loss/acre.

As an example, a loss of approximately 600 bed feet of either onions or garlic at plant populations of 20 (garlic) - 22 (onions) plants per bed foot can result in losses in the range of 1,000 (garlic) - 2,000 (onions) pounds per acre. At 1983 costs, this could result in a per acre loss in onions of \$80.00/acre and in garlic of \$100.00/acre. Losses in this range have been observed on an annual basis in both garlic and onion fields, and for either crop can mean the difference between profit and loss for the grower.

Revised
2. 1983
1983 and 1984

though the total
was about 100
existing trees

following form
step from which

Number of
estimated
trees
1983

with a loss of approximately 100 trees of 100 years or
a total population of 200 (100 trees) - 100 trees
result in loss in the range of 100 (100 trees) - 100 trees
acre. At 1983 a loss of 100 trees in a 100 acre loss
of 200,000 acres, loss of 100 trees
loss in both years and
difference between the

Page 2

Abstract - Geographic Distribution and Economic Importance

For both garlic and onions there is the additional loss of harvestable product to the processor, a loss in quality due to the potential for a percentage of decayed bulbs being processed with good bulbs, plus the loss and cost of seed. For garlic, seed costs range from \$700.00 to 800.00/acre while for onion they are generally lower in the range of \$40.00 to \$100.00/acre.

Losses in garlic have been reduced through the use of soil sampling to determine *S. cepivorum* sclerotia population levels prior to planting and in-furrow treatments with Botran. Future losses are expected to be even further reduced with the expected registration of Rovral.

Distribution of White Rot of Onions in Ontario

P. Oudemans and L.V. Edgington

Ontario has approximately 1715 hectares (4,237 acres) worth 12.7 million Canadian \$ in onions as reported in 1981. Onions are primarily grown in muck soils in three regions; (1) Grand-Bend-Thedford, (2) Leamington and (3) the Holland-Colbar-Keswick marshes. White rot has been present in the Grand-Bend-Thedford marsh for more than six years but no exact estimate of disease loss has been attempted.

The largest muck area (3) producing 70 percent of the onions in Ontario, was first observed to be infested in 1979. Since then 6 farms widely distributed within this region have developed white rot. We estimate on the farm where the disease has been known since 1979, 5 hectares are infested with ca. 5% of plants diseased. Approximately 30 percent of this marsh is in onions. Current rotation practices are with onions every two to three years.

DISTRIBUTION AND ECONOMIC IMPORTANCE OF 'WHITE ROT' OF
ONIONS, IN AUSTRALIA

Prudence Somerville and Peter Merriman

Department of Plant Pathology, University of California, Davis, CA, and
Plant Research Institute, Burnley, Victoria, 3121, Australia, respectively.

White rot was first reported in Australia in 1928 in the Melbourne market garden district. Since then it has spread to the other onion-growing areas of Victoria and onion acreage has declined dramatically. In New South Wales and in South Australia, the major producer of white and brown onions, outbreaks of 'white rot' have been sporadic and confined to less than 20 acres. Recent outbreaks in South Australia however suggest the disease could become more prevalent in commercial production. In Queensland and Tasmania the disease is economically significant. In Queensland 10 percent of the onion acreage is affected and in Tasmania 24 outbreaks have been reported in the last three years and the industry is threatened.

AVERAGE PRODUCTION OF WHITE & BROWN ONIONS IN AUSTRALIA -
 1980-1982
 (AUSTRALIAN BUREAU OF STATISTICS)

STATE	AREA (HECTARES)	PRODUCTION (TONS)	YIELD (TONS/HA)
SOUTH AUSTRALIA	1,167	34,892	29.9
QUEENSLAND	796	24,428	30.7
NEW SOUTH WALES	581	19,465	33.5
TASMANIA	498	19,179	38.5
WESTERN AUSTRALIA	292	15,094	51.7
VICTORIA	643	14,391	22.4

THE DISTRIBUTION OF ALLIUM WHITE ROT IN FINLAND

In Finland the most commonly grown onion species are Allium cepa and Allium cepa var. aggregatum.

A. cepa is grown in the southern and central parts of Finland and A. cepa var. aggregatum in northern Finland. Each year the total area used for growing onion crops is more than 700 hectares.

A. cepa is grown from sets mostly imported from other European countries. Seed is used in Southwestern Finland only. Farmers who produce A. cepa var. aggregatum use sets which have grown in their own fields.

Other important Allium-species are leek and garlic, which are both cultivated in southern Finland.

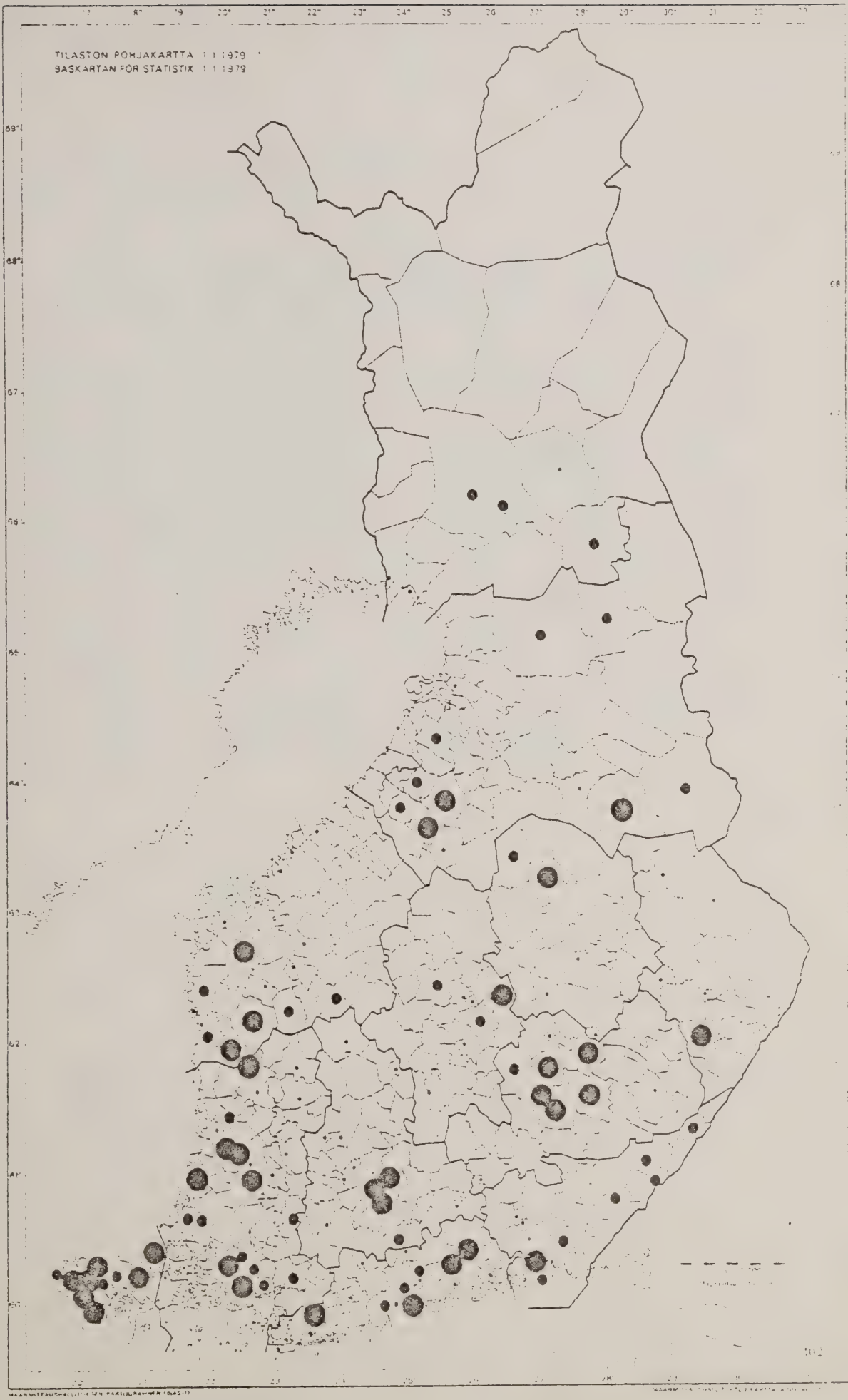
Onion white rot has been found on all of the above mentioned Allium-species except on A. cepa var. aggregatum.

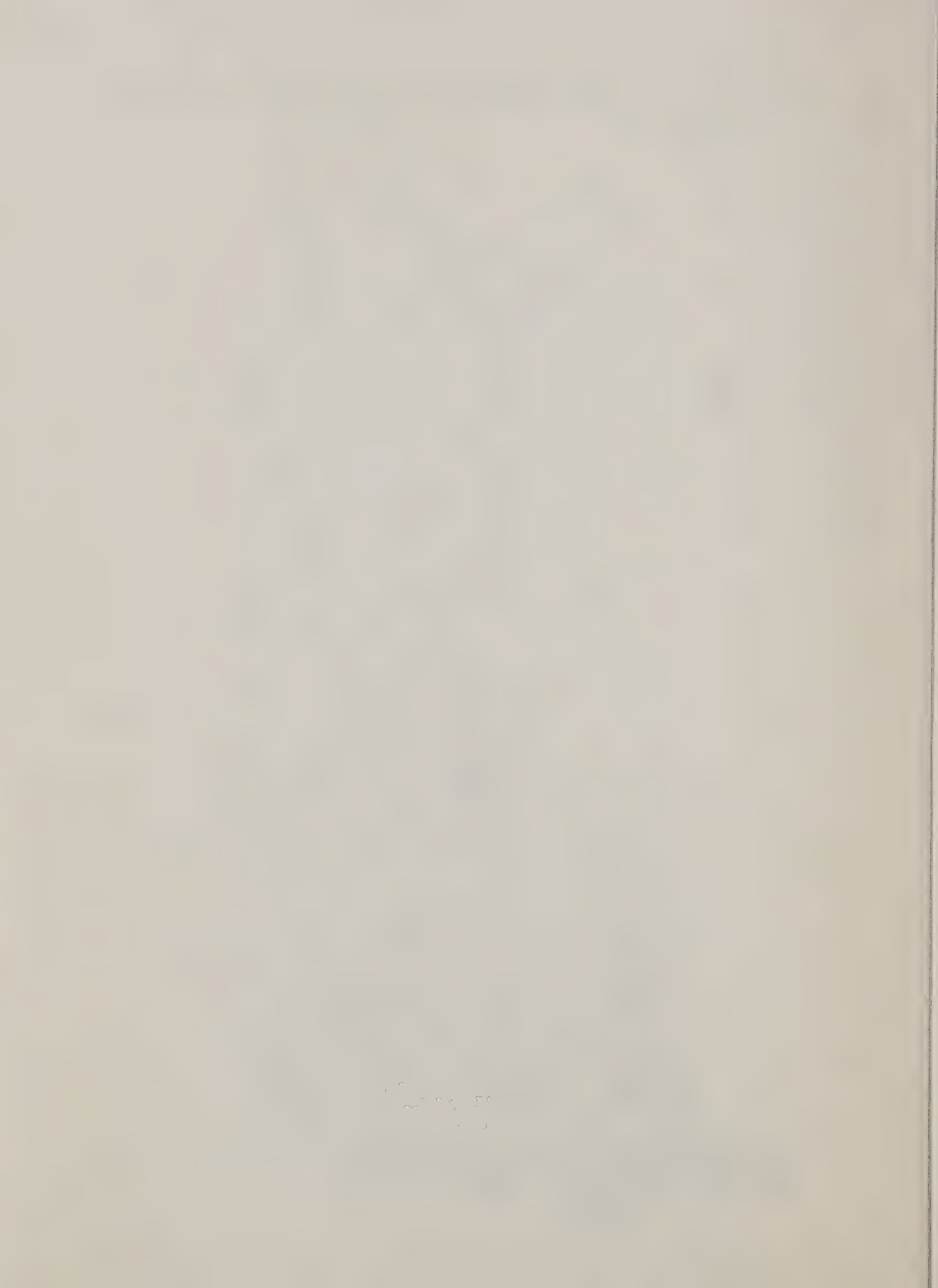
The map shows the municipalities where white rot was found in the years 1968-1982.

The big spots are municipalities where white rot appears, and the small ones are municipalities from where onion samples were collected, but the disease was not found. We can see here that white rot is spread over the most important onion production areas in Finland.

Onion yield losses change from year to year. In our experiments, losses have been between 4 and 76 percent of the yield. Some growers have even reduced onion crops because of the big yield losses due to white rot.

At present, we can say that white rot is a big problem in Finland. Laws have now been established for measures to be taken when the disease is found on onions and other Allium-species. But, in addition to these measures, good methods for the control of white rot are needed. I hope this conference will be of great help regarding this matter.





THE ECONOMIC IMPORTANCE OF ALLIUM WHITE ROT IN ITALY

A. GARIBALDI

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White rot, caused by Sclerotium cepivorum, was recorded on garlic for the first time in northern Italy in 1901. It has since been regarded as a common and sometimes destructive disease of onion and garlic in several regions of the Country. Actually it can cause severe damages to winter onions produced in the Central and Southern parts of the country and on garlic produced during autumn and winter in the cooler garlic producing areas (Piedmont, Liguria). The severity on leek seems generally lower than on other Allium. The percentage of infected bulbs and corms is very variable: from 0 to 70-80 % in some vegetable-producing areas, where the growers produce green onions for fresh consumption. In many regions it is necessary to control the white rot by dusting onion plantlets and garlic corms at transplanting time or by applying fungicides to the soils during sowing.

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Second International Workshop on Allium white rot

Session III: Allium white rot: Geographic distribution and economic importance

N. I. Georgy

Onion is one of Egypt's most important crops. Egyptian onions are pungent, very firm and, when cured properly, keep well. The onions can be handled without injury and can be shipped for long distances under unfavourable conditions. Egyptian onions are greatly desired for their high quality and early appearance in foreign markets - over 80% of the exports are shipped in from March to June.

Cultivars: There are three main onion cultivars. The first (cv Behairy syn. Giza 20) is cultivated in the northern Delta region. Most of the Delta region is still free from white rot infestation. The second cultivar is Improved Giza 6, which is suitable for the more southerly Upper and Middle regions. About 60% of these areas are now contaminated by the pathogen (Sclerotium cepivorum). The third cultivar, Shandawil 1 is new and matures early. Cv. Shandawil, also suitable for growing in Upper and Middle Egypt, can be harvested during December and January thus escaping the critical periods of infection by S. cepivorum. This latter work has been done mainly by the Onion Breeding Section under the direction of Dr. El Gammal.

Cultivation: Egyptian onions are cultivated in three different ways.

Transplants: In Middle and Upper Egypt, seeds of Giza 6 are sown in nursery beds in mid August-September and the seedlings transplanted 60-70 days later in October - November. Thirty kg seed is needed to sow 1 feddan (4200 m²) of nursery bed and the seedlings from one feddan are sufficient for 6 - 8 feddan of transplanted crop depending on spacing. Bulbs are harvested the following March.

Onion sets: Dr. El Gammal from Egypt who is attending the 2nd International Workshop on Allium white rot will discuss the details of this method.

Direct seedings: Some attempts are being made to plant seeds directly to their final positions. The small size of many fields, however, would not help the introduction of mechanisation. Fields are larger on the new lands which may facilitate mechanisation.

Crop areas: Onions cultivated as a single crop as in Upper and Middle Egypt.

The areas and yields of onions grown as a single crop in Upper and Middle Egypt are shown for the years 1960-65, before the building of the Aswan High Dam and the subsequent spread of white rot, compared with the last five seasons (Table 1). There has been a sharp decrease in the area cultivated and in yields, mostly (c 70%) due to white rot. This has resulted in a progressive reduction in exports.

Onions intercropped with cotton amount to 70-150,000 feddan, and are used for local consumption.

- 2 -

White rot situation in Egypt. At the First Workshop, Dr. El Moity from Egypt described the first appearance of white rot in 1929 and the work of Dr. Mikhail in 1964 on the reasons for its spread north and south along the Nile Valley.

Since then, the pathogen has appeared in new areas. The author recorded white rot in a small area at Garbia Governorate (Delta region) in 1979. Since then the decision has been taken that this area must not be cultivated with any sort of onion crop.

Dr Mikhail (1964) recorded in his Bulletin that white rot had not been found in the Fayoom Governorate in Middle Egypt. This area is an oasis naturally isolated from the Nile Valley by a 15-km barrier of desert. The appearance of white rot was recorded five years ago in Tamia (Fayoom Governorate). Bulbs introduced from Upper Egypt for seed production were the main source of this new infection. This has resulted in a sharp decrease in the onion area and in yields.

In Egypt, we need to find a practical solution for controlling this serious disease and thus return to the golden age of the Egyptian onion and restore the good reputation of Egyptian onions in foreign markets.

In this direction we work in three ways viz, with chemicals, with biological control agents, and with integrated control by using early maturing set varieties or sets treated with the best fungicide, the optimum rate of fertilisation (NPK) and optimum dates of sowing.

We hope we can find the good solutions in the near future.

TABLE 1

Total onion area (feddan) and the yield (Ton) in
Middle and Upper Egypt

Year	Middle Egypt			Upper Egypt			Total area (Feddan)	Total yield (Ton)
	Beni- swif	Minia	Fayoom	Asuit	Sohag	Kena		
1960	4448	12274	1169	3108	11013	1237	33248	233101
61	3983	9190	1224	3111	10729	1114	29351	180284
62	5728	10584	1947	4608	11860	1101	35828	237632
63	5916	14720	2719	5390	14770	1247	44762	353963
64	6094	17149	1822	4410	12023	1015	42513	327409
65	5505	12267	1531	4552	15924	762	40541	326535
78	3545	1234	4315	1869	10470	896	22329	187628
79	443	1122	3988	1726	4248	53	11580	118515
80	876	1179	5829	1093	4648	183	13808	122331
81	1061	806	4532	669	3915	128	10108	98649
82	833	402	3713	502	2067	62	7579	63955

Here I have chosen the three Governorates from Middle and Upper Egypt which account for the biggest export. Observe how the total area and yield has decreased.

SECOND INTERNATIONAL WORKSHOP ON ALLIUM WHITE ROTIII Allium White Rot: Geographic distribution and Economic ImportanceAllium white rot in the U.K.

A R Entwistle

The Allium crop areas and values in 1982 compared with 1979 are as follows:

	1979		1982	
	ha	£1000s	ha	£1000s
Salad onions				
- summer	967		1284	
- overwintered	912	12333	992	14378
Dry bulb				
- summer	6407		5722	
- overwintered	1875	20179	1139	17204
Leeks	1742	8277	1892	9244

In 1980-82 crops were badly affected by white rot during spring and summer. In 1981 and 1982, warm temperatures in autumn resulted in above average infection late in the season. In spring 1983, the onset of white rot has been delayed by about a month. This is apparently due to a combination of low temperatures and of high rainfall. Observations at the NVRS indicate that the wet conditions have resulted in soil compaction which has impeded white rot development. Where soils have been loosened, infection has been rampant. Autumn-sown, overwintered crops frequently became infected in spring. Whilst control measures with iprodione are generally successful in salad crops, they are not reliable in overwintered bulb crops. This may be because improvements in application methods are needed, as treatment is frequently delayed until the onset of symptoms.

White rot is becoming more of a problem in leeks. Increasingly, direct drilling is replacing transplanting. Growers with expertise in growing salad onions also grow high quality close-spaced leeks for the supermarket trade. On these farms, rotations between Allium crops can be small with an attendant risk of white rot and an urgent need for reliable control measures. Stem base treatment with iprodione is generally used in leeks.

IV Chemical Control

INTRODUCTION: Needs and outlook for chemical control
A R Entwistle

Iprodione (Rovral 50% w.p.; May & Baker plc), applied as a combined seed and stem base treatment is the officially approved method for the control of white rot in UK salad crops (Entwistle & Munasinghe, 1980a).

The treatment is in extensive use and provides major benefits to the grower by reducing white rot losses and, as a consequence, improving land usage on the farm (Birch, 1979).

The benefits to other Allium crops, however, have not been so marked and clearly further improvements are needed. Improvements may be sought with

- a) different methods of application
- b) fungicides with greater activity
- c) fungicides from different chemical groups.

Application. Maximum effectiveness with iprodione is achieved by a combination of pre- and post-drilling treatments. Traditionally in the UK, pre-drilling treatment has been by seed treatment. Where seed drilling rates are high as in salad onions (10-30 kg/ha), fungicide placement is high and control is effective. Linking the fungicide placement with seed drilling rates, however, has the disadvantage that with low seed rate crops such as direct drilled leeks and dry bulb onions (3-5 kg seed/ha) there is unlikely to be adequate protection. Increasing the fungicide loading on seeds in compensation for low seed rates is impractical because of the dual problems of drilling and of phytotoxicity. Therefore other methods of applying pre-drilling treatments are needed. Furrow treatments, either as sprays (Adams, 1978), as granular formulations (Entwistle & Munasinghe, 1980b) or by gel-incorporation as in fluid-drilled crops (Entwistle & Munasinghe, 1981) have all shown promise experimentally, but have not been adopted in commerce. Furrow treatment might also be of benefit in non-seeded crops, eg. transplanted leeks (UK) and onions (Egypt) and onion crops grown from sets and garlic crops grown from cloves. Post-drilling treatment is by band spray applied 5 wk from drilling in summer crops, in the spring following autumn sowings (Entwistle & Munasinghe, 1980a) or at the onset of symptoms. Stem base treatments depend on accuracy of placement for their effectiveness and in the UK only a single bed is treated with each pass of the sprayer. Applying the sprays at the same time of drilling would improve accuracy because the sprays would be aligned with the seed drill. For this modification to be effective, some account would have to be taken of the leaching and persistence characteristics of the fungicide.

It should be possible to dispense with stem base treatments altogether, eg. by replacing them with a pre-drilling furrow treatment in which fungicide is also positioned in the surface layers of the soil with stem base treatments.

Activity. A constraint of present treatments is their action in preventing rather than curing symptoms. With the high rates of fungicide needed there is an understandable reluctance to treat crops which may subsequently prove not

to be at risk. Conversely, even land with no previous history of Allium cropping, and therefore considered to be safe from attack, can be a source of severe infection. Deciding which crops to treat is thus something of a gamble.

There would be clear advantages in a treatment which would be reliably effective when applied at the onset of symptoms. Control measures could then be directed where they are most needed.

Unfortunately, iprodione appears to be insufficiently active when used in this manner (Entwistle & Munasinghe, 1980a) and more recent results with vinclozolin have been similar (Entwistle & Marian, 1983a). The results with meclozolin at the NVRS (UK) in 1982, however, showed considerable promise (Entwistle & Marian, 1983a). Meclozolin is currently restricted to experimental use.

Future screening experiments for fungicides should therefore include a test of eradicant as well as of protectant activity.

Chemical structure. Dicarboximide fungicides are currently the most effective against white rot in the UK and are the most commonly used. Such reliance on chemicals from a common chemical group has the risk - yet to be evaluated in white rot - of the development of fungicide resistance (sensu Dekker, 1982) in S. cepivorum.

Iprodione resistance in S. cepivorum occurs readily in vitro (T. Jeves, personal communication, 1980). The possibility that in vitro resistance reflects a similar occurrence in field conditions needs urgent investigation. In vitro cross-resistance in S. cepivorum, between vinclozolin, iprodione and meclozolin has been shown in the author's laboratory (Entwistle, 1983, unpublished data). Furthermore, cross-resistance between dicarboximide fungicides and dicloran, has been shown in Botrytis (Leroux et al., 1979).

The use of fungicides with differing chemical structures is one strategy of reducing the risk of fungicide resistance at least in air-borne pathogens. For reasons of economy, the alternating use of different fungicides may be preferable to the use of mixtures (Skylakakis, 1981; Jeffery & Kable, 1982).

Outlook. In 1981/82, iprodione failed to control white rot at the NVRS (Entwistle & Marian, 1983a) and on two commercial farms. Extensive enquiries confirmed that these were isolated occurrences. The cause of this lack of control is still under investigation. Tests at NVRS have failed to demonstrate dicarboximide resistance in S. cepivorum from infected, iprodione-treated plants (Entwistle, 1983b).

Vinclozolin, another dicarboximide fungicide, gave good control in such conditions - evidence that dicarboximide resistance was not a factor. These studies are continuing.

In overwintered onions 1982/83, stem base treatment with iprodione in spring 1983 (manufacturers' recommendation) gave better control of the spring phase of the disease than one-third the rate applied in autumn. Iprodione also failed to control white rot the previous autumn.

Table 1. Comparison of different iprodione treatments on white rot incidence in salad onions, NVRS 1982/83a.

Iprodione ^b applied to:		% white rot	
seed	stem base	15 November 1982	5 May 1983
62.5	0.15 in March '83	46	1
50	0.05 in September '82	50	30
Untreated		74	-

^a Crop sown on 10 August 1982; ^b Rate: seed - g a.i./kg seed; stem base - g a.i. in 100 ml water/m row.

One explanation might be that iprodione has been leached from the surface layers of the soil following above average rainfall in the UK. Furthermore, there are unconfirmed reports in the UK that iprodione gives a poorer standard of control on light sandy soils, possibly due to leaching.

Studies are now urgently needed to clarify the causes of the failures so that the future potential can be determined of dicarboximides for the control of white rot.

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Margins

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Use of Metham to Control Allium White Rot

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In laboratory experiments, columns of soil containing sclerotia of Sclerotium cepivorum Berk. were drenched with solutions of various concentrations of metham (Vapam®, 32.7% sodium-N-methyl dithiocarbamate). After four days, the sclerotia were isolated from soil, surface sterilized, placed on acidified potato-dextrose agar containing rose bengal (33 g/ml) and incubated at 20 C for 14 days to determine their viability. Metham killed 50% of the sclerotia at 16-17 µg a.i./ml and 95% at 50 µg a.i./ml.

In three New Jersey production fields containing sandy soils, metham was injected into sprinkler irrigation lines (chemigation) at a rate of 234 L product/ha (25 gal/A) over a 2-3 hr irrigation period. The length of the irrigation period was adjusted to apply approximately 2.5 cm (1 in) of irrigation water. The concentration of metham in the irrigation water was about 350 µg a.i./ml. These treatments were applied 4-5 weeks before planting the Allium crops. Analysis of soil samples collected from the fields 1-2 days after

1 treatment application indicated that the metham treatment killed 83-100% of the
2 S. cepivorum sclerotia.

3 In late July, 1981 leek seedlings (A. porrum L.) were transplanted into one
4 field. Beltsville bunching onions (A. cepa x A. fistulosum amphidiploid) were
5 directly seeded in a second field in September, 1981. The third field was not
6 planted. At harvest, about May 1, 1982, there were approximately 4 times as
7 many plants in the metham treated plots than in the untreated control plots
8 (Table 1). White rot was significantly reduced in both fields even though 70%
9 of the leek plants exhibited signs and symptoms of white rot. The lack of
10 disease control in the leek field was attributed to the substantial rainfall
11 (3.1 cm) the night of the metham application and to the nonuniform irrigation
12 treatment (0.6 - 5.6 cm). There was a substantial increase in yield of both
13 leeks and bunching onions in the field tests which more than paid for the cost
14 of the treatment (Table 1).

5 In addition to white rot control, metham provided significant control of
6 weeds in the treated plots (2). Based on these results and those obtained
7 previously (1,2) farmers are now using this method of control for Allium white
8 rot and for a number of soilborne pests.

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Table 1. The effect of metham applied through sprinkler irrigation at 234 L/ha on incidence of *Allium* white rot, crop yield, and value of yield

Crop	Treatment	Percent		Estimated		Value of yield/ha ^a	Increased profit/ha ^b
		Total no. of plants/plot	marketable plants/plot	Percent white rot	yield (crates/ha)		
Leeks	Control	120	8	100	153	\$ 981	--
	Metham	147	51	70	1,082	6,936	\$ 5,572
Bunching onions	Control	203	60	49	897	4,449	--
	Metham	877	97	3	7,598	\$37,686	\$32,854

^aValues are based on the average price of \$6.41 for leeks and \$4.96 for bunching onions per crate received by farmers in southern New Jersey in 1981.

^bValues were determined on the value of the metham treated yield minus the value of the yield from the control, minus the cost of the metham (\$383/ha).

IV Chemical control of Allium white rot at NVRS, UK, 1981-83

A R ENTWISTLE

Experiments at NVRS have tested the effects of 1) iprodione, 2) vinclozolin and meclozolin, 3) thiabendazole and 4) carboxin, oxycarboxin, tolclofos-methyl, and triadimefon.

Iprodione. Contrary to previous experience, iprodione failed to control white rot at the NVRS in 1981-82 (Entwistle & Munasinghe, 1980; Entwistle & Marian, 1983a). The effect was first observed in spring 1982 in overwintered salad onions (Table 1). These onions were growing on land used three years previously for testing fungicides and which had been fallowed in the intervening period. Subsequently experiments sown in April 1982 gave similar results (Table 1).

Table 1. Changes in effectiveness of iprodione on control of Allium white rot, NVRS.

Iprodione ^a applied to		% white rot
seed	stem base	
1. Sown 21 April 1978		10 October '78
62.5	0.0625 on 26 May	6
Untreated		88
2. Sown 12 August 1981		13 May '82
50	0.05 on 12 October	46
Untreated		100
3. Sown 20 April 1982		27 July
50	0.05 on 25 May	85
Untreated		99
4. Sown 16 July 1982		19 October
50	0.2)	54
50	0.05) on 20 August	90
Untreated		75

The causes for this change in response are not known. A new sample of iprodione gave similar results. However, a four-fold increase in active content of the stem base treatment in 1983 reduced infection by half (Table 1) but this was still very much less than in previous years. Extensive tests for iprodione resistance in S. cepivorum have proved negative (Entwistle, 1983c). In one experiment, iprodione gave proportionally better control on plots which had not been previously treated with iprodione, compared with plots treated three years previously (Entwistle, 1983b).

Vinclozolin. A combined seed and stem base treatment with vinclozolin effectively controlled white rot in overwintered salad onions (Table 2) (Entwistle, 1983a). Similar results were obtained in spring - 1982-sown salad onions. In this experiment, seed treatment at 50g vinclozolin/kg seed combined with a stem base treatment of 0.05g vinclozolin/m row (total rate 3 kg vinclozolin/ha) controlled the disease for 20 wk, whereas at half this rate the period of control was 15 wk. A stem base treatment applied 5 wk after drilling to plants grown from untreated seed was ineffective.

Table 2. Effect of vinclozolin on white rot incidence in salad onions
NVRs, 1981-82.

20

Vinclozolin ^a applied to		% white rot	
seed	stem base	13 May '82	10 June
1. Sown 12 August '81			
50	0.05)	1	0
25	0.025) on 12 October '81.	3	13
Untreated		100	100
2. Sown 24 May 1982			
50	0.05)	<1	7
25	0.025) on 29 June '82	3	25
0	0.05)	52	52
Untreated		97	-

Meclozolin. When first tested in 1982, a seed treatment at 50g meclozolin/kg seed combined with a stem base treatment at 0.05g a.i./m row (total rate 3 kg meclozolin /ha) controlled white rot for 27 wk (Table 3) (Entwistle, 1983a). Good results were also obtained at half the rate.

Table 3. Effect of meclozolin on white rot incidence in salad onions, NVRs 1982.

Meclozolin ^a applied to		% white rot	
seed	stem base	27 July	28 October
Sown 20 April			
50	0.025	0	2
Untreated		99	99

The effect of meclozolin was also tested on white rot already present in the crop. Meclozolin was applied as a stem base treatment 9 wk after drilling to plants grown from untreated seed. At that time 27% plants had white rot. White rot declined in meclozolin-treated plants and continued to increase in untreated plants (Table 4).

Table 4. Effect of meclozolin on infection in salad onions, NVRs 1982.

Meclozolin stem base treatment ^{a,b}	% white rot	
	30 July	22 October
	2	0
Untreated	62	100

^b Sown 22 April.

Thiabendazole. Thiabendazole is applied as a seed treatment and marketed in the UK for the control of white rot under the trade name Bromotex Onion Seed Dressing (abbreviated to Bromotex O.S.D.). Thiabendazole is applied at 25g a.i./kg seed together with an inert carrier forming a pellet around the seed. This product was tested at NVRs in 1982.

Thiabendazole at 25g/kg either as pure compound or as Bromotex reduced white rot at 11 wk but not at 15 wk (Table 5). Pure thiabendazole applied at 150g a.i./kg seed had more effect (Entwistle & Marian, 1983c).

Table 5. Effect of thiabendazole on Allium white rot, NVRS 1982.

Thiabendazole seed treatment ^{a,b}	% white rot	
	6 July	28 July
25 (Bromotex OSD)	13	96
25 (NVRS)	30	90
150 (NVRS)	4	29
Untreated	68	83

^b Sown 15 April 1982.

Other fungicides. None of the remaining fungicides tested was effective at controlling white rot (Table 6) (Entwistle & Marian, 1983b).

Table 6. Effect of various fungicides on Allium white rot, NVRS 1982.

Fungicide applied to [*]	total kg/ha	% white rot	
		7 July	28 July
furrow ^b	stem base		
1. Carboxin			
0.2	0.2 (3) ^c	24phytotoxic.....
0.05	0.05 (3) ^c	6	41 87
2. Oxycarboxin			
0.02	0.6 (1) ^d	24	10 58
0.05	0.15 (1) ^d	6	73 97
3. Triadimefon			
0.05	0.15 (1) ^d	6phytotoxic.....
4. Tolclofos-methyl			
0.05	0.05(2) ^e	4.5	47 77
5. Untreated	-	36	91

^{*} Sown 20 April

^b Rate: furrow - g a.i./m row; stem base - g a.i. in 100 ml water/m row/treatment on ^c6 / 19 May, 2 June^d 25 May^e, 29 June.

^a Rate: Seed treatment - g a.i./kg seed; stem base treatment - g a.i./ in row.

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SECOND INTERNATIONAL WORKSHOP ON ALLIUM WHITE ROT

IV Chemical control

The significance of iprodione-sensitivity in S. cepivorum

A R Entwistle

In spring 1982, iprodione-treated onions became infected with white rot at NVRS. This contrasted with previous experience (Entwistle & Munasinghe, 1981; Entwistle & Marian, 1983). A similar loss of control occurred on two commercial farms. To the best of the author's knowledge, however, these were isolated occurrences.

First, isolates from sclerotia in soil and on onions on these farms (termed Farm isolates) were compared for their in vitro response to iprodione with isolates known to be sensitive to 5 ppm iprodione (termed NVRS isolates) and with in vitro induced iprodione-resistant isolates (supplied by T Jeves, Dept of Botany, The Polytechnic, Liverpool, UK). Only induced iprodione-resistant isolates grew in the presence of 5 ppm iprodione (termed I); all isolates grew in a similar time on fungicide-free malt extract medium (termed FFM). It did not appear therefore that fungicide-resistance was the cause of the loss of control.

There is the possibility, however, that in vitro response to fungicide may not reflect the ability of the fungicide to control disease. Therefore the effects of iprodione and of vinclozolin on white rot incidence were tested in field plots inoculated with two Farm isolates and compared with the effects in similar plots inoculated with two NVRS isolates. Iprodione and vinclozolin were applied as a seed treatment at 50 g a.i./kg seed followed 5 wk after drilling with a stem base treatment at 0.05 g a.i./m row (total rate 3 kg a.i./ha). The experiment was sown in August 1982 and overwintered.

There was no difference in disease control with either type of isolate (Table 1).

Infected iprodione- and vinclozolin-treated onions were tested for dicarboximide resistance by culturing mycelial fragments, sclerotia and roots on dicarboximide medium.

In November 1982, S. cepivorum was isolated within 2-4 d on FFM from 14 iprodione-treated plants from NVRS plots and from 6 similar onions from Farm plots. There was no growth on I. Isolations were made onto iprodione medium only (I).

In May 1983, isolations from mycelial fragments were also tested in the presence of 5 ppm vinclozolin and meclozolin (termed V and M respectively) using a modified form of the technique used for testing sclerotia (Entwistle, 1983). Isolations from iprodione-treated onions failed to grow on VIM by 9 d. Isolations from vinclozolin-treated onions from Farm plots yielded S. cepivorum in 3-5 d on VIM. Whatever dicarboximide medium S. cepivorum was first isolated on, there was cross-resistance to the other dicarboximides.

Table 1. Effect of iprodione and vinclozolin on S. cepivorum infection with two sources of isolate

	Nos of healthy and infected plants/0.5 m row			
	23 November 1982		24 May 1983	
	healthy	infected	healthy	infected
<u>NVRS isolates</u>				
untreated	3	9	4	7
iprodione	43	45	4	36
vinclozolin	48	0	34	18
<u>Farm isolates</u>				
untreated	9	15	2	13
iprodione	34	27	13	54
vinclozolin	60	0	62	14

It was apparent, however, that colony appearance, the time to start growth and growth rate differed according to the source of the isolate.

Isolates from FFM grow within 2 d on fresh FFM, compared with 4 d on dicarboximide medium. Isolates from V, I M took 2 d to start growth on all media. Colony size at 4 d was less on VIM than on FFM when the isolates originated from FFM. The converse was evident when isolates originated from VIM and colonies also had a dark central region (c. 30-50% of colony area); this change in colour was also seen in two isolates from V and M when subcultured on V.

DISCUSSION

S. cepivorum can readily become VIM resistant in vitro (T Jeves, personal communication, 1980). Throughout a number of experiments at the NVRS, the following range of responses was observed.

1. No growth
2. Growth limited to point of inoculation
 - hyphae turn dark
 - hyphae proliferate into an irregular, sclerotial-like mass in which normal-looking sclerotia form. These sclerotia grew on FFM and are VIM-sensitive.
 - sclerotia placed on VIM proliferate in a similar manner as hyphae
 - hyphae and sclerotia form a tuft of white hyphae which grows away from the surface of the medium. These hyphae are VIM-sensitive and the tuft is apparently a reaction against the dicarboximide (see also Keyworth & Milne, 1969).
3. Growth into the dicarboximide medium
 - dark, slow-growing colonies (c. 0.1 cm/d)
 - dark colonies sector into normal colonies
 - normal colonies form directly (2-31 d).

VIM-resistance can occur almost immediately (3-5 d) or after a period of apparent inactivity (1 month). In every instance of VIM-resistance in isolations from the field at NVRS, there has been a delay in the time at which growth starts on VIM compared with FFM. When transferred to fresh VIM, there is no delay and growth occurs at 1000 ppm.

Similar results were obtained with a range of isolates, some held in stock culture since 1975, prior to the field use of dicarboximide fungicides.

Although there is no evidence that loss of control at the NVRS is due to VIM-resistance, studies are needed to determine the role of dicarboximide instability in the field.

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VINCLOZOLIN ABSTRACT

Vinclozolin has been tested against Allium-white rot on onion and garlic. Several application methods have been assessed alone and in various combination. Satisfactory results have been obtained in regard to efficacy and yield increase. Best overall performance was achieved with seed/bulb dressings in combination with post directed spray applications. Crop safety has generally been good with all methods tested even when auxiliary materials (methyl cellulose, dextrose) were used to improve adherence of vinclozolin on seed.

The following methods of application, rates and timings were tested:

A. Onions/Direct Seeding

1. seed dressing - 25-37.5 grams ai/kg of seed
2. spray application
 - infurrow at planting - 0.05% ai into the furrow before closing
 - broadcast - 1.0-1.5 kg ai/ha 4-6 weeks postemergence and 6-8 weeks before harvest
 - post directed (banded 5-10 cm) - 0.5% ai 4-6 weeks postemergence stem base treatment
 - soil drench - 0.05% ai/0.5 liter per meter of row 4-6 weeks postemergence

B. Onions/Transplanted or Planted as Bulbs

1. dipping treatment - 0.2% ai/15 min

C. Garlic

1. clove dressing - 1.0-1.5 grams ai/kg cloves
2. infurrow at planting - 0.05%-0.10% into the furrow before closing

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Six years of field trials in the Salinas Valley and at Tulalake, California have been conducted on the control of white rot on onion and garlic. A number of approaches have been made. In summary the data indicate the following:

Garlic:

1. Application of fungicides into the planting furrow at time of planting in a 3" band as the garlic "seed" is dropped has been the most successful and economic method of control. The following fungicides have been successful:

Dichloran	75% W.P.	@ 56.94	g/100 meters of row
Iprodione	50% W.P.	@ 22.78	" " " "
Vinclozolin	50% W.P.	@ 22.78	" " " "
2. Combinations of fungicides worked well but results were not consistently superior to those obtained with single materials.
3. Treatment of garlic "seed" with amounts of fungicide equal to that placed in the furrow resulted in ~~infusion~~ control when compared with in-furrow application. *inferior*
4. Methyl bromide injected 6" deep into the soil under 1 ml polyethylene tarp at 350 Kg/ha resulted in almost complete control. Chloropicrin at the same rate resulted in minimum control.
5. Vapam applied through the sprinkler system in 1.75" of water at 456 l/ha (50 gal/A) resulted in minimum reduction of viable sclerotia and no economic control of disease.
6. Level of control with fungicides is directly related to inoculum levels in the soil. Our current thinking is that when sclerotial levels reach above 5-6/100 g of soil it would be advisable to plant some crop other than garlic.

Onions:

Work with onions has resulted in less conclusive data. Under our warmer summer growing conditions the occurrence of severe white rot disease is quite erratic and it has led to many trials with data of little value. In addition, in-furrow applications of fungicides such as is done in garlic has been a difficult thing to accomplish because the very narrow knife like planting shoe used by our growers does not lend itself to a good band application of fungicide.

We have experimented with fungicide application 3/4" below the seed line through the use of horizontal blade but the results have not been encouraging.

Blade application of iprodione at time of planting plus a topical application after emergence has proved ineffectual.

Pelleting of seed with iprodione @ 300 g/Kg seed has resulted in consistent reduction of white rot losses. However, this method has not consistently provided economic control.

Session IV. Chemical Control

Practical Results 1979-1983

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Fungicide trials were conducted from 1979-1983 in a field (soil type: Aura Sandy Loam, 56-32-12% sand-silt-clay) which was infested with *Sclerotium cepivorum*-infested oat grain inoculum in the fall of 1976 and cropped continuously with onions each winter. Bunching onions, cv. Beltsville Bunching, were used in all tests and seeded with a "Planet Jr." mechanical planter in rows spaced 0.6 meters apart in early September for each test. Fungicides were applied in the 1979 and 1980 tests as in-furrow sprays with a CO₂-pressurized sprayer that had a nozzle placed immediately behind the planter but prior to the covering shoe. For the 1981, 1982 and 1983 tests, fungicides were applied as a preemergence spray after seeding. For some treatments a single spring application was made in mid-March with a tractor-mounted boom sprayer equipped with 3 nozzles spaced 0.5 meters apart that delivered 421 l/ha at 40 psi.

Fungicides evaluated included iprodione (Rovral 50W) at 0.6 and 1.12 kg a.i./ha, vinclozolin (Ronilan 50W) at 0.6, 0.8 and 1.12 kg a.i./ha, Serinal 50W at 0.8 and 1.12 kg a.i./ha, dicloran (Botran 75W) at 8 and 34 kg a.i./ha, and benomyl (Benlate 50W) at 1.12, 3.6 and 7.1 kg a.i./ha. Iprodione, vinclozolin and Serinal provided a reduction in incidence of white rot, Table 1. In the majority of tests, the addition of a spring application resulted in improved control. The preemergence application of fungicide was as effective as the in-furrow application. In-furrow applications were difficult to accomplish because the nozzle pressure resulted in blowing the seed out of the furrow in many instances.

In New Jersey, control of white rot is accomplished by the use of crop rotation where possible, soil fumigation or the use of iprodione at planting. Soil fumigation is performed by soil injection of a multipurpose chemical such as methylisothiocyanate (280 l/ha) or the addition of metham in the irrigation system (234-467 l/ha). For the 1982-1983 overwintered crop, an emergency exemption registration was in effect in New Jersey for the use of iprodione at 1.12 kg a.i./ha at seeding.

TABLE 1. Control of onion white rot with a fall and spring application of fungicide, Bridgeton, N.J.

Treatment & Rate kg a.i./ha	% White Rot			
	1980	1981	1982	1983
Iprodione - 0.6	3	0	12	-
Iprodione - 1.1	0	18	22	33
Vinclozolin - 0.6	7	-	14	-
Vinclozolin - 0.8	-	14	10	-
Vinclozolin - 1.1	-	-	12	31
Control	47	68	38	43

SECOND INTERNATIONAL WORKSHOP ON ALLIUM WHITE ROT

ABSTRACT - CHEMICAL CONTROL

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Trials conducted in California on onions and garlic by Dennis Hall, Art Greathead, Fred Crowe, Prudence Somerville and the author from 1978-83 have involved numerous chemicals and methods of application.

The following list is provided to indicate what types of application techniques have been tested on the indicated crops:

GARLIC

1. Pre-plant fumigation
2. Clove treatment
3. In-furrow at planting

ONION

1. Pre-plant fumigation
2. Bed top shank injection
3. Bed top incorporation
4. Pre-plant band treatment (simulated in-furrow)
5. Pre-plant spray blade
6. In-furrow at planting
7. Coated seed

The data relating to chemicals applied by these various techniques and the degree of disease control obtained will be provided by Art Greathead.

LABORATORY RESISTANCE IN SCLEROTIUM CEPIVORUM BERK.
TO DICARBOXIMIDES

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White rot, caused by Sclerotium cepivorum Berk., is a serious disease of onion and other Allium species in Italy. Among the different fungicides, dicarboximides (iprodione, procymidone, vin clozolin) provide a very good control of the pathogen applied as a combi nation of plant dusting at transplanting and of soil drenching during growing period.

Considering the risk of appearance of resistance to dicarboximides, as observed in the case of other pathogens (i.e. Botrytis cinerea, Monilinia spp., Alternaria spp.) and the fact that resistance of S. cepivorum to PCNB, fungicide chemically related to dicarboximides, was earlier detected in vitro (Garibaldi, 1966), it seemed interesting to evaluate the possibility of selecting in vitro strains of the pathogen resistant to dicarboximides.

Strains of S. cepivorum resistant to dicarboximides were easily selected under laboratory conditions, even if at different frequency from various strains, varying between 2 and 12 % of the tested mycelium disks.

The resistant strains obtained in vitro were slightly vi rulent compared with the sensitive parent strains. Dicarboximide-resis-
tant strains of S. cepivorum showed cross-resistance to all the fungici-
des belonging to this group and to dicloran and PCNB. All the tested strains were sensitive to benomyl.

The resistant strains selected in vitro, showing a high level of resistance and a low virulence do not seem able to give practical problems; but the risk of appearance of strains with a lower level

2.

of resistance, virulent, and able to survive in field conditions and to compete with the sensitive strains, as observed in the case of B. cinerea, should not be underestimated.

Specific Tolerance of Sclerotium cepivorum to Dicarboximide Fungicides

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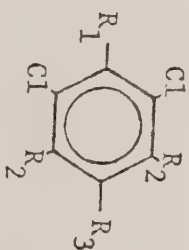
(Manuscript submitted to PLANT DISEASE)

ABSTRACT

Five isolates of Sclerotium cepivorum tolerant to one dicarboximide fungicide showed cross-tolerance to the dicarboximides iprodione, myclobutin and vinclozolin, and to dichloran and PCNB, but not to benomyl, captan or thiram (Table 2). The dicarboximides, dichloran and PCNB share a common structural subunit (Table 1). Dicarboximide EC_{90} values for most of the tolerant isolates were >1000-fold those of the parent isolates (Table 3). Sensitivity to benomyl was unchanged by tolerance to dicarboximides (Table 2). Wide variations in the sensitivities of 14 dicarboximide-sensitive isolates of S. cepivorum to PCNB and thiram were noted (Table 2). Frequency of occurrence of tolerance to dicarboximide fungicides was too low and variable to obtain a reliable estimate (Table 4).

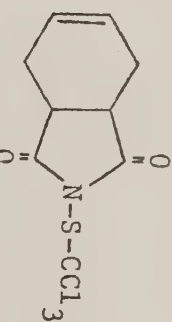
Table 1. Structural relationships among eight fungicides tested for toxicity to Sclerotium cepivorum

Fungicides containing the structural subunit



N-substituted 3,5-dichlorophenyl

Structurally unrelated fungicides



Captan

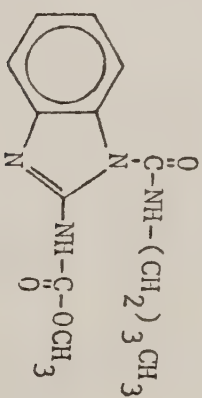
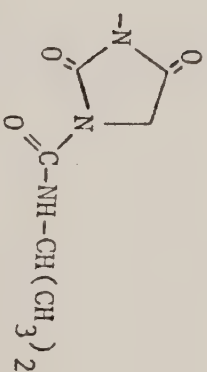
Fungicide	R ₁	R ₂	R ₃
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PCNB	-Cl	-Cl	-N ₂ O
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Dichloran	-NH ₂	-H	-N ₂ O
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Iprodione

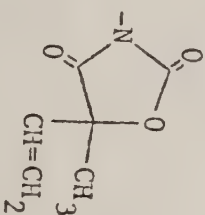
-H -H



Benomyl

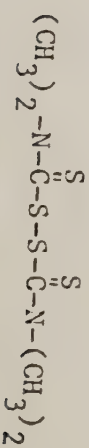
Vinclozolin

-H -H



Meclozolin

-H -H



Thiram

Table 2. Highest concentrations (ppm) of various fungicides in potato dextrose agar allowing detectable mycelial growth of various isolates of Sclerotium cepivorum

Isolate	Source	Vinclozolin	Iprodione	Meclozolin	Dichloran	PCNB	Benomyl	Thiram	Captan
'S' Isolates									
J191	Hull, UK	0.1	0.1	0.1	0.1	0.01	1	10	1000
NZ32	Auckland, New Zealand	1	1	0.1	0.1	0.01	1	100	1000
BBY	Burnaby, B.C.	0.1	0.1	0.1	0.1	0.01	1	1000	1000
ELS	Fairview, Alberta	0.1	0.1	0.1	0.1	0.01	1	1000	1000
GF	Grand Forks, B.C.	1	1	0.1	0.1	0.01	1	1000	1000
NZ37	Auckland, New Zealand	0.1	0.1	0.1	0.1	1	1	10	1000
S187a	Australia	0.1	0.1	0.1	0.1	100	1	10	1000
S197a	Australia	0.1	0.1	0.1	0.1	100	1	10	1000
S197b	Australia	0.1	0.1	0.1	0.1	100	1	10	1000
S201a	Australia	0.1	0.1	0.1	0.1	100	1	10	1000
J192	Hull, UK	0.1	0.1	0.1	0.1	100	1	100	1000
VDM	Wageningen, Holland	0.1	0.1	0.1	0.1	100	0.1	100	1000
S187b	Australia	1	0.1	0.1	0.1	100	1	100	100
S201b	Australia	0.1	0.1	0.1	0.1	100	1	100	1000
'T' Isolates									
S201bv	S201b	1000	1	10	1000	1000	1	1000	1000
S187bVa*	S187b	100	10	10	1000	1000	1	1000	1000
S187bv	S187b	1000	1000	10	1000	1000	1	10	1000
J191v	J191	1000	1000	1000	1000	1000	1	100	1000
VDM1	VDM	1000	1000	1000	1000	1000	0.1	1000	1000

* Reverted to sensitivity after 25 transfers on unamended potato dextrose agar.

Table 3. Comparative^a EC₉₀ values for vinclozolin and iprodione against radial growth of tolerant (T) and parental (S) isolates of Sclerotium cepivorum on fungicide-amended potato dextrose agar

Isolate	Status	EC ₉₀ (ppm)	Isolate	Status	EC ₉₀ (ppm)
Vinclozolin					
S187b	S	0.39	S187bV	T	>1000
S201b	S	0.38	S201bV	T	>1000
J191	S	0.38	J191V	T	>1000
VDM	S	0.43	VDMI	T	>1000
Iprodione					
S187b	S	0.52	S187bV	T	>1000
S201b	S	0.35	S201bV	T	<10 ^b
J191	S	0.44	J191	T	>1000
VDM	S	0.40	VDMI	T	>1000

^a $r^2 > 0.933$ for all regressions; $P \leq 0.025$.

^b Precise value not determined.

Table 4. Frequency of occurrence of tolerant sclerotia of Sclerotium cepivorum

		<u>Isolate</u>		
	<u>VDM</u>	<u>S187b</u>	<u>S201b</u>	<u>J191</u>
<u>Vinclozolin</u>				
Replications	10	12	15	14
No. Tested	9828	9040	14497	15059
No. Germinated	21	6	4	4
Mean Frequency	1/468	1/1507	1/3624	1/3765
<u>Iprodione</u>				
Replications	7	9	10	11
No. Tested	4877	7466	11643	12084
No. Germinated	7	2	3	1
Mean Frequency	1/525	1/2063	1/3743	1/12084
<u>Untreated</u>				
Replications	21	23	25	19
Mean Germination	88.1 %	70.7 %	89.0 %	96.1 %

REVIEW OF LITERATURE WITH RESPECT TO SCREENING OF ALLIUM FOR RESISTANCE TO WHITE ROT.

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SCREENING METHODS

In the Netherlands very probably the degree of white rot attack in a certain contaminated field varies widely over a series of years. However no data are available on this.

Addition of inoculum to a field can increase strongly the percentage of diseased plants but in the Netherlands such an addition, resulting in 300 sclerotia per kg of (clay-)soil, did not give any remarkable white rot attack.

Very probalby the following factors (can) have strong influence on the percentage of diseased plants in the open:

- TEMPERATURE. Rather cool temperatures (15° - 20°C) seem to be optimal for white rot development (WALKER, 1924, 1926; LOCKE, 1966; ADAMS and PAPAIVIZAS, 1971; ENTWISTLE and MUNASINGHE, 1978; UTKHEDE and RAHE, 1978; CROWE and HALL, 1980; COLEY-SMITH et al., 1981).
- INOCULUM POTENTIAL. In literature mention is made of 1-10 sclerotia per kg of soil as causing slight attacks and 10-100 as causing very severe attacks (TICHELAAR, 1965; CROWE et al., 1980) in the open. For screening purposes under field conditions mostly much higher concentrations are used, e.g.: 60 sclerotia per 2 seeds (UTKHEDE and RAHE, 1978), or about 100.000 sclerotia per m² (VALDIVIA, 1975).
- PATHOGENICITY. Mentioning is made of differences in pathogenicity between local strains of Sclerotium cepivorum (WALKER, 1926; TIMS, 1948; COLEY-SMITH and HOLT, 1966; ADAMS and PAPAIVIZAS, 1971; ABDELRAZIK, 1975; UTKHEDE et al., 1978). However very little or nothing is known of differences in pathogenicity between local strains under natural conditions.
- MYCOSTATIC EFFECT. This factor is indicated by SCOTT (1956) and COLEY-SMITH (1968) but probably up to now little is known of the background of such effects. This factor could cause the failure of field screening in the Netherlands.

Although very interesting results have been obtained by field screening (VALDIVIA, 1971; UTKHEDE and RAHE, 1980) the above mentioned (and other) factors makes this type of screening less reliable. Therefore screening (of seedlings) under controlled conditions seems more attractive. Under such conditions all the 4 above mentioned (and other) factors can be kept constant, e.g. a temperature of about 15°C, a standardized substrate with little or no mycostatic effect and a constant number of sclerotia, of a pathogenic isolate, per seedling.

Such screening methods have been developed by ADAMS and PAPAIVIZAS (1971), UTKHEDE and RAHE (1978) and UTKHEDE et al. (1978).

COLEY-SMITH (1981) has developed an interesting screening method using as a criterion the germination of sclerotia in the presence of seedlings.

Very essential for the efficiency of screening under controlled conditions is a high correlation level between the results of such screening and those of field screening. UTKHEDE and RAHE (1978) did find such a (significant) correlation amounting (only) 0,26.

RESULTS OF SCREENING

Probably one of the first descriptions of screening for white rot resistance is that of NATTRASS (1927). Screening of 6 cultivars was done in a heavily infected field. The best cultivar was Lemon Rocco (4.3% infection) and the most susceptible White Welsh (29% infection). On the other hand HOLMES SMITH (1930) mentions strong resistance of White Welsh.

ADAMS and PAPAIVIZAS (1971) screened some onion cultivars and 16 *Allium* species under controlled conditions. Only *A. caeruleum* showed remarkable resistance.

After screening of 4 varieties in the open VALDIVIA (1971) found Red Creole being the most resistant.

SEMB et al. (1976) screened 45 cultivars in infected soil without finding significant differences. Of 10 cultivars grown from sets, Rijnsburger was the best. In an other trial including 15 cultivars, Rizzi was the best (46% healthy) and Rijnsburger was the second-best (36% healthy). Screening of several *Allium* species proved some of them to be immune (*A. obliquum*, *A. aflatunense* and *A. schoenoprasum*) and one to be strongly resistant (*A. tuberosum*). However in the trials of ADAMS and PAPAIVIZAS (1971) *A. schoenoprasum* and *A. tuberosum* proved to be susceptible. Also COLEY-SMITH (1959) mentions susceptibility of *A. schoenoprasum*.

ALI (1976) found that Zittauer had the best resistance after screening of 13 cultivars.

RONDOMANSKI and DORUCHOWSKI (1976) found some resistance in the cultivar Rawska, but this resistance was not durable.

DHYANI and CHAUHAN (1976) mention Nasik Red as a resistant variety.

UTKHEDE and RAHE published a number of articles describing the screening of a large number of onions accessions most of them being PI numbers. Screening was done under field conditions (with and without added inoculum) and under controlled conditions. Their most interesting results are contained in an article of 1980, in which Ailsa Craig is mentioned as the most resistant cultivar (41% diseased plants as an average of 5 trials) and Canada Maple as a very susceptible cultivar (69% diseased plants as an average of 5 trials).

COLEY SMITH et al. (1981) did not find differences between onion cultivars with respect to stimulation of the germination of sclerotia. *A.tuberosum* also strongly stimulated the germination (see under SEMB et al.). Little or no stimulation was observed from *A.stipitatum*, *A.aflatunense*, *A.christophii* and *A.karataviense*. Strangely enough an experienced Dutch grower of ornamental *Allium* species supposes that *A.stipitatum* (as well as *A.cowanii*) is very resistant to white rot and *A. christophii* is very susceptible.

CONCLUSIONS AND SUGGESTIONS

- Resistance to white rot found so far is not very impressive. Looking for higher levels of resistance is very urgent.
- The inheritance of the resistance found so far is not yet known. Screening is not very eliminative and results of the screening of off-springs of surviving plants have not yet been published.
- Up to now little attention has been paid to international exchange of resistant material. This situation should be improved.
- One or two cultivars should be used as international standard(s) for screening on white rot resistance in the seedling stage under controlled conditions. Pukekohe Longkeeper and Beth Alpha might be very suitable cultivars for this purpose.
- Knowledge is necessary of the correlation between the results of screening seedlings under controlled conditions and those of screening under field conditions.
- More knowledge is wanted of the pathogenicity of *Sclerotium cepivorum* strains under field conditions.
- More knowledge of mycostatic effects in different soil types is also essential.

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SCREENING FOR RESISTANCE TO WHITE ROT CAUSED BY SLCEROTIUM CEPIVORUM BERK.
IN ONIONS (ALLIUM CEPA L.) AND LEEKS (ALLIUM PORRUM L.)

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METHODS AND MATERIALS

Isolates of *Sclerotium cepivorum* are maintained on potatoe dextrose agar (PDA) in a refrigerator at about 4°C.

For large scale production sclerotia from former mass cultures are disinfected by shaking them during 1-2 minutes in a 1% solution of NaClO. Afterwards they are washed in sterile destilled water and dried on filter paper whereafter they are placed separately on PDA at 22°C. About a week later well growing pure cultures are used for the initiation of the sclerotia production on a barley medium. A very high yield is realized within 20 days at about 20°C. Sclerotia are separated by blending and washing in water. Finally they are dried on filter paper.

The barley substrate is prepared as follows: 130 ml ammoniumtartrate solution (3 g/l) is added to 100 g of barley seeds. This mixture is autoclaved at 120°C during one hour on two successive days.

For the sake of standardization a large bulk of sclerotia is produced from a pathogenic isolate. This bulk is kept at -20°C and sclerotia from it are used for all screening work to be done.

Some investigations were done on the inoculum potential. Some results are given in Tables 1 and 2 of the following paragraph. Such results together with experience from practice (in respect of the occurrence of white rot and of the growing of onion seedlings in a greenhouse) were utilized to standardize the screening method as follows:

Seeds are sown in pots (diameter 4.5 cm) with standard peat soil (Trio) mixed with 25% sand. In each pot one hole is made by using a pencil and in each hole 3 seeds are sown, together with a 'spoon' full of sclerotia. This 'spoon' is an iron bar in which a little hole has been drilled. One scoop contains about 200 sclerotia. Per cultivar 8 pots are used for screening and 8 other pots (also with 3 seeds each) as controls.

The seedlings are grown at 14°C in a greenhouse of the phytotron. Additional light (one H PI/T lamp of 400 Watt per m² at 1 m above the soil surface) is given during daytime (about 9 hours) in November, December, January and February.

The majority of the inoculated plants of susceptible cultivars die within 7 weeks after sowing. Surviving plants are kept in a greenhouse at 10°C. Some more plants die back at this temperature.

At the beginning of bolting the remaining plants are transferred from 10°C for multiplication.

After the development of the above described method 300 accessions of the IVT onion collection and 93 accessions of the IVT leek collection were screened for resistance. The Rijnsburger strain Jumbo was used as a standard. Some data representing the best resistance found so far are shown in Tables 3 and 4.

RESULTS

Table 1. Numbers of surviving seedlings of two onion cultivars after exposure during seven weeks to different quantities of sclerotia of Sclerotium cepivorum. Spring 1982.

Quantity of sclerotia per planting hole	Rijnsburger strain Jumbo			Cultivar Beth Alpha		
	Repl.1	Repl.2	Repl.3	Repl.1	Repl.2	Repl.3
Control	18	17	9	18	19	20
1 spoon of sclerotia*	0	0	0	0	2	2
2 spoons of sclerotia	0	0	0	0	2	1
3 spoons of sclerotia	0	0	0	0	4	0
4 spoons of sclerotia	0	0	0	1	1	0

* ca. 200 sclerotia

Table 2. Numbers of surviving seedlings of two onion cultivars after exposure during five weeks to six levels of three isolates of Sclerotium cepivorum. Autumn 1982.

Quantity of sclerotia per planting hole	Rijnsburger strain Jumbo			Cultivar Beth Alpha		
	Isolate 1	Isolate 2	Isolate 3	Isolate 1	Isolate 2	Isolate 3
Control	26	26	27	37	38	29
50 sclerotia	5	20	4	12	29	19
1 spoon of sclerotia ¹⁾	1	0	2	3	14	7
2 spoons of sclerotia	0	3	0	2	2	5
4 spoons of sclerotia	1	0	2	3	1	6
8 spoons of sclerotia	0	0	1	2	2	2

¹⁾ ca. 200 sclerotia

Table 3. Numbers of surviving seedlings of three onion cultivars seven weeks after inoculation with Sclerotium cepivorum sclerotia. Summer 1982.

Cultivar		Repl.1*	Repl.2	Repl.3	Repl.4	Repl.5	Sum	Surviving seedlings as % of control
Jumbo	control	11	13	14	17	15	70	
	inoculated	3	0	2	1	5	11	16
Beth Alpha	control	16	23	20	19	21	99	
	inoculated	8	3	9	9	10	39	39
Pukekohe Long-keeper	control	17	18	21	21	16	93	
	inoculated	7	6	16	16	11	46	49

*For each replicate 8 x 3 seeds were used.

Table 4. Numbers of surviving seedlings of six leek cultivars and one onion cultivar seven weeks after inoculation with Sclerotium cepivorum sclerotia. Spring 1983.

Crop	Cultivars	Origin	Treatment	Repl.1*	Repl.2	Repl.3	Repl.4	Repl.5	Sum	Surviving seedlings as % of control
				Repl.1*	Repl.2	Repl.3	Repl.4	Repl.5	Sum	control
Leeks	Batina	Netherlands	control	12	15	13	18	10	68	
			inoculated	2	1	4	2	6	15	22
"	Carentan	Denmark	control	21	17	20	19	21	98	
			inoculated	0	2	8	5	3	18	18
"	Eleplant	Denmark	control	19	17	12	15	9	72	
			inoculated	4	1	6	3	3	17	24
Onions	Jumbo	Netherlands	control	23	22	16	23	21	105	
			inoculated	1	0	1	1	6	9	9
Leeks	Zwitserse Reuzen	Netherlands	control	8	7	8	8	11	42	
			inoculated	0	0	0	2	0	2	5
"	Odin	Denmark	control	15	17	16	13	13	74	
			inoculated	1	0	1	0	0	2	3
"	Artica	Netherlands	control	17	14	14	15	12	72	
			inoculated	1	0	0	1	3	5	7

*For each replicate 8 x 3 seeds were used.

The Nature of Resistance in Allium Species and Cultivars.

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Introduction.

There are only a few reports of Sclerotium cepivorum infecting plants other than members of the genus Allium. Mukerji (1970) recorded what he considered to be S. cepivorum infecting Belamcanda chinensis (Iridaceae) as well as several species of Zephyranthes (Amaryllidaceae) and Young & Allen (1969) were able to infect seedlings of flax (Linum usitatissimum), white clover (Trifolium repens), cabbage (Brassica oleracea var capitata) and tomato (Lycopersicon esculentum) under aseptic conditions. The results of Young & Allen have led Rahe (1981) to suggest that the specificity of S. cepivorum for Allium is not due to tissue resistance of non-host species but to soil fungistasis (Coley-Smith et al., 1967) acting in conjunction with specific stimulation of germination of sclerotia by Allium species (Coley-Smith, 1960; Coley-Smith & Holt, 1966).

Within the genus Allium there is little sign of resistance to S. cepivorum. Adams & Papavizas (1971) tested 16 species and only found any evidence of resistance in A. caeruleum. With the methods which they used however it would not have been possible to distinguish resistance based upon inability to stimulate germination of sclerotia from true tissue resistance.

Amongst onion cultivars evidence has now been produced (Utkhede & Rahe, 1978a,b; Utkhede et al., 1982) that resistance does occur but its level is low and can only be detected in experiments done on a large scale. Rahe (1981) considers that resistance amongst onion cultivars observed in field trials may result from differential stimulation of sclerotium germination by resistant and susceptible cultivars and is probably expressed prior to the establishment of a pathogen mycelium - host tissue interface. There is at present no direct evidence that onion cultivars differ in their ability to stimulate germination of sclerotia.

In view of this lack of direct evidence we have started to re-examine the basis of resistance in non-host and host species and cultivars.

1. Non-Allium Species

Non-Allium species have been tested using the nylon strip soil-tube method

(Coley-Smith, 1960; Esler & Coley-Smith, 1983). Germination of sclerotia was induced by treating the tubes with garlic extract. Seedlings growing aseptically on water agar were also tested by inoculating their roots either with a mycelial disc of S. cepivorum or with sclerotia which had been pre-germinated on agar. The results for a few of the species tested are shown in Table 1. In soil, infection was confined to members of the Liliaceae, although not all of these became diseased even when germinating sclerotia were in contact with their roots. Browning of the roots of many species was seen indicating that resistance reactions had occurred. Under aseptic conditions a wider range of species became infected but much less infection was seen where sclerotia were used than where the inoculum consisted of mycelial discs. The results indicate that except in a few members of the Liliaceae resistance in non-Allium species is controlled by failure to stimulate germination of sclerotia and by tissue resistance factors. Under aseptic conditions this tissue resistance can be overcome, particularly if the inoculum potential is high, but this is hardly surprising with a necrotrophic fungus like S. cepivorum. It is interesting that species of Zephyranthes did not become infected in soil and all attempts to infect them under unsterile conditions have proved fruitless.

2. Allium species

A large number of Allium species have been tested for their capacity to stimulate germination of sclerotia (Esler & Coley-Smith, 1983). The majority of species were highly stimulatory but six ornamental species either failed to stimulate germination altogether or only had a weak ability to do so. The reason for this is that the non-stimulatory species contained S-methyl-L-cysteine sulphoxide as their principal flavour and odour precursor and had low overall flavour and odour levels. The stimulatory species contained either S-1 or S-2-propenyl-L-cysteine sulphoxide as their major flavour and odour precursor and with a few exceptions their overall flavour and odour levels were high.

The non-stimulatory Allium species have now been tested for susceptibility to S. cepivorum using the nylon strip soil-tube method with sclerotia stimulated

to germinate by the addition of garlic extract (Table 2).

It is quite clear that the roots of all Allium species are susceptible to infection and that lack of infection within the genus is controlled by pre-infection factors.

3. Edible Allium species

Although it is commonly believed that garlic cultivars are particularly susceptible to white rot and that leek cultivars are more resistant than onion cultivars there have been no systematic trials to investigate such differences. During 1980 and 1981 two trials were completed on severely infested plots to investigate possible differences in resistance. In 1980 all the cultivars except that of garlic were grown from seed (Table 3). The outstanding features of the results were the low levels of infection in all four leek cultivars and the high level of infection in the garlic cultivar. The only onion cultivar with a low level of infection was Rynsburger Waldo in which seedling emergence was very poor. The low level of infection in the widely spaced plants of this cultivar suggested that variation in spacing might give some information about whether infections were mostly primary, from germinating sclerotia, or also secondary from plant to plant spread. In the 1981 trial all plants were grown singly in peat blocks, the distances between blocks being 4.4 cm (close spacing), 8.8 cm (medium spacing) and 13.2 cm (wide spacing). The results (Table 4) again showed low levels of infection in the leek cultivars and a very high level of infection in the two garlic cultivars. There was very little effect of spacing suggesting that the majority of infections resulted from germinating sclerotia.

Since 1981 most of our work has been concerned with the differences between the stimulatory capacity of onions and leeks. A difficulty with the soil-tube germination technique, when using a small number of replicates, is that variation between replicates is considerable and this variation can obscure small differences in the stimulatory capacity of species or cultivars. Experiments have therefore been done on a larger scale. Fig.1 shows the results from a soil tube experiment with seven onion and seven leek cultivars. In this experiment for each cultivar

there were 10 replicate tubes each containing 20 sclerotia giving a total of over 70,000 observations over the course of the 24 wks. of the experiment. Individual germination percentages at 14 wks are shown in Table 5. We also did a field experiment in 1982 in which sclerotia in nylon bags were placed under plants grown in multiseeded peat blocks, 5 plants per block. The results are shown in Fig.2. There was little difference between onion, garlic and A. fistulosum cultivars but the response of sclerotia under leeks was much slower.

It is clear that the lower levels of infection seen in leek compared with onion cultivars result at least partly from their weaker capacity to stimulate germination of sclerotia. At present we are investigating the reasons for the differences in stimulatory capacity. Since flavour and odour compounds are known to be involved we have examined these in a large number of cultivars (Table 6). In spite of previous reports (Freeman & Whenham, 1975) the methyl content of leeks, at least at the end of the season, is very similar to that of onions. Flavour strength is higher, on a dry weight basis, in onions than in leeks but this is obviously associated with the high % dry matter content of the latter. Analyses of roots are now being made throughout the growing season but so far no major differences between onions and leeks have been detected and the reasons for the difference in the stimulatory capacity of these two species remains a mystery.

4. Onion cultivars

Although there is no direct evidence that onion cultivars differ in stimulatory activity a programme of work designed to detect possible differences has been started. Stimulatory activity is known to be associated with flavour and odour compounds and these have been examined in 100 or so cultivars. No evidence has been found to indicate any differences in flavour and odour type and attention is now focussed on analyses of flavour strength.

Several tests have been made with a number of Egyptian cultivars known to be badly affected by white rot and said to be strong flavoured. End-of-season analyses (Table 7) have shown that they are stronger flavoured than most European cultivars when grown under UK conditions. They do however grow very poorly in

the U.K. We have also done some soil tube germination experiments with them and have demonstrated that the seedlings of these Egyptian cultivars are more stimulatory than those of European cultivars (Fig.3). Attempts to do field stimulation experiments with Egyptian cultivars have proved fruitless because of the poor growth of the latter.

Our attention is now being concentrated on a number of American cultivars which under UK conditions show extremes of flavour levels. These are being tested in the field for both stimulatory activity and susceptibility to white rot. No results from these experiments are available at present.

The fact that flavour and odour levels can affect stimulatory capacity has been shown by growing plants of Rijnsburger Robusta under different levels of sulphur nutrition in washed sand. This resulted in extremes of flavour and odour much greater than those recorded between cultivars. A clear relationship between the stimulatory activity of extracts made from these plants and their flavour and odour strength has been demonstrated.

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Table 1Reaction of non-Allium species to S. cepivorum

Species	Family	Root infection		
		Soil tubes	Petri dishes Mycelial Sclerotia disc	
Brassica oleracea var capitata	Cruciferae	-	+++	+
Linum usitatissimum	Linaceae	-	++	-
Lycopersicon esculentum	Solanaceae	-	+++	+
Trifolium repens	Leguminosae	-	++	-
Zephyranthes candida	Amaryllidaceae	-	++	-
Zephyranthes grandiflora	"	-	++	-
Ipheion uniflorum	Liliaceae	++	+++	+++
Nectaroscordum siculum	"	+++	NT	NT
Nothoscordum inodorum	"	++	NT	NT
Tulbaghia violacea	"	+++	+++	+++
Allium cepa	"	+++	+++	+++

+++ = severe, ++ = moderate, + = slight

NT = not tested.

Table 2

Reaction of Allium species to germinating sclerotia of S. cepivorum in soil.

Species	% germination of sclerotia ^a	Sclerotia formed on roots or bulbs ^b	Root infection seen after clearing ^b
A. aflatunense	0	+	+
A. caeruleum	56	-	+
A. cepa	84	+	+
A. cristophii	0	+	+
A. cyaneum	48	-	+
A. karataviense	0	+	+
A. oreophilum	NT	+	+
A. rosenbachianum	8	+	+
A. stipitatum	6	+	+
Distilled water control	3		

NT = not tested

a = tubes treated with distilled water, b = treated with garlic extract.

Allium fistulosum by white rot disease (Sclerotium cepivorum).

Species	Type	Cultivar	Seedling emergence %	Plants with white rot ^a
<u>Allium cepa</u> (onion)	Dry bulb	Ailsa Craig	43	56.6
		Bedfordshire Champion	49	44.5
		Giant Rocca Brown	59	45.8
		Giant Zittau	53	53.9
		Hydeal	70	43.6
		Hygro	77	46.3
		James Long Keeping	51	50.7
		Reliance (Oakey)	54	57.6
		Red Italian	52	54.3
		Rijnsburger Augusta	57	52.8
		Balstora	68	47.5
		Jumbo	61	45.8
		Robusta	40	46.3
		Waldo	23	27.4
		Royal Oak	55	58.5
<u>A. porrum</u> (leek)	Bunching	White Spanish	52	46.0
		White Lisbon	47	48.5
	Pickling	Paris	50	55.1
<u>A. sativum</u> (garlic)		Giant Winter (Inverno)	78	18.1
		Musselburgh	72	14.4
		Gennevilliers Splendid	67	15.7
		Titan	60	16.2
<u>A. fistulosum</u>		Rose Duvar	91	82.6
		Asagi Kyoto Market	69	43.7

^a Percentages transformed to arcsin valuesLSD between two treatment means : 11.2 ($P \leq 0.05$); 19.1 ($P \leq 0.01$)

Table 4 Percentage white rot (Sclerotium cepivorum) infection in cultivars of onion, leek, garlic and Allium fistulosum ^a

Species	Cultivar	Spacing			Species X cultivar means	Species means
		Close	Medium	Wide		
<u>Allium cepa</u> (onion)	Rijnsburger Robusta	76.7	72.9	64.5	72.0	75.1
	White Spanish	79.9	78.1	76.3	78.1	
<u>A. porrum</u> (leek)	Musselburgh	12.1	23.3	21.2	18.9	17.3
	Gennevilliers Splendid	9.7	18.2	19.5	15.8	
<u>A. sativum</u> (garlic)	Rose Duvar	88.2	90.0	87.3	88.5	89.0
	Brignoles 4	88.5	90.0	90.0	89.5	
<u>A. fistulosum</u>	Ishikuro	54.4	59.4	54.6	56.2	55.5
	Asagi Kyoto Market	51.9	57.7	55.2	54.9	
Spacing means		57.8	61.2	58.6		

^a Percentages transformed to arcsin values

LSDs between means: spacing, 2.7 ($P \leq 0.05$); species X cultivar, 4.3 ($P \leq 0.05$), 5.6 ($P \leq 0.01$); species, 4.0 ($P \leq 0.01$)

Fig. 1. Soil tube sclerotium germination experiment

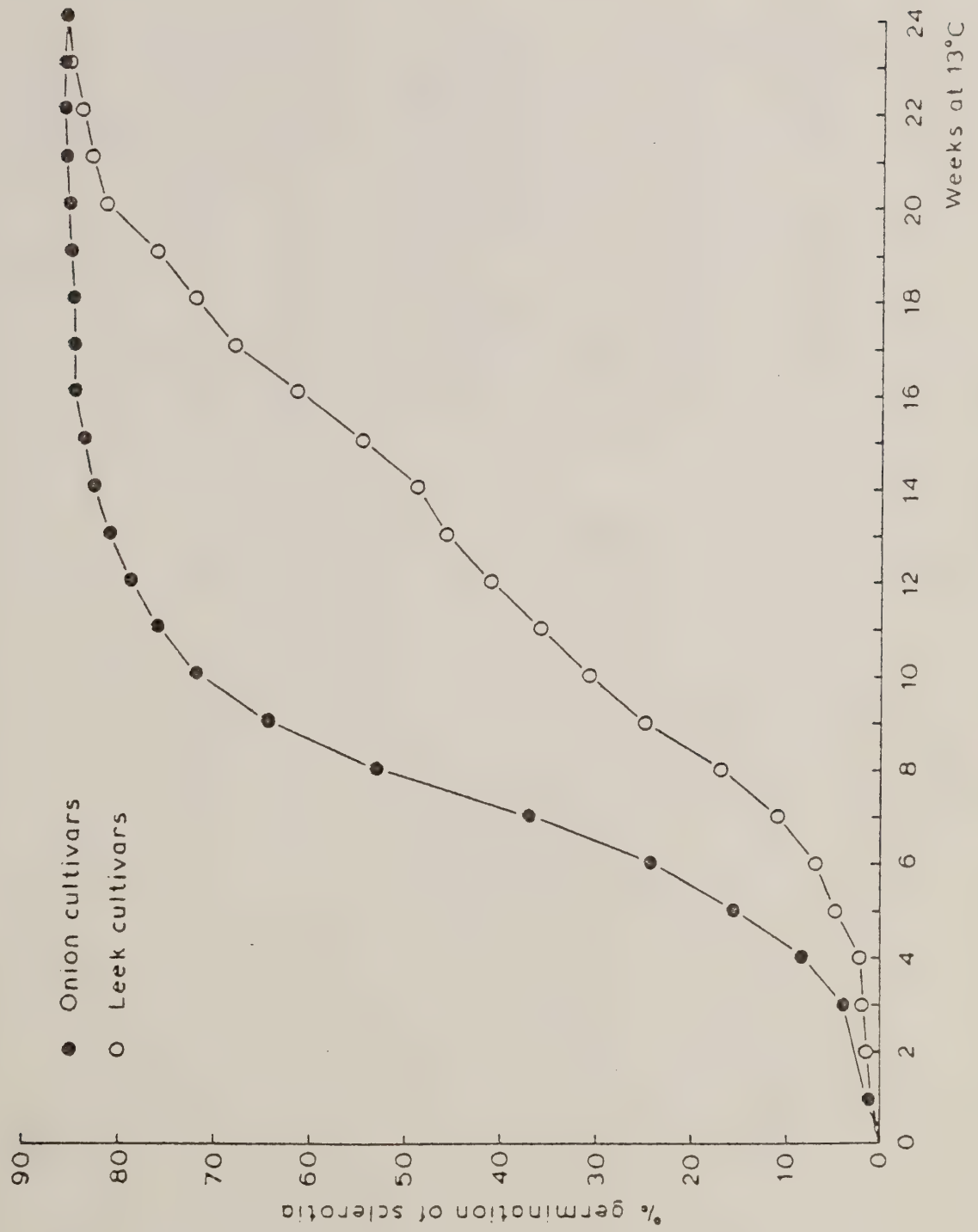


Table 5

Percentage germination of sclerotia (J11) after 14 weeks at 13°C in soil tube experiments.

Leek	Exp.1	Exp.2	Onion	Exp.1	Exp.2
Musselburgh	47	25	White Lisbon	89	89
Walton Mammoth	47	62	Rijnsburger	86	82
Giant Winter	49	45	Bedfordshire Champion	83	81
Lyon	38	27	Ailsa Craig	80	88
Inverno	61	68	Jumbo	65	80
Thor	55	53	Robusta	74	84
Odin	59	65	Valencia	88	80
Mean	61	49	Mean	81	81

Fig. 2. Field sclerotium germination experiment (Isolate J412)

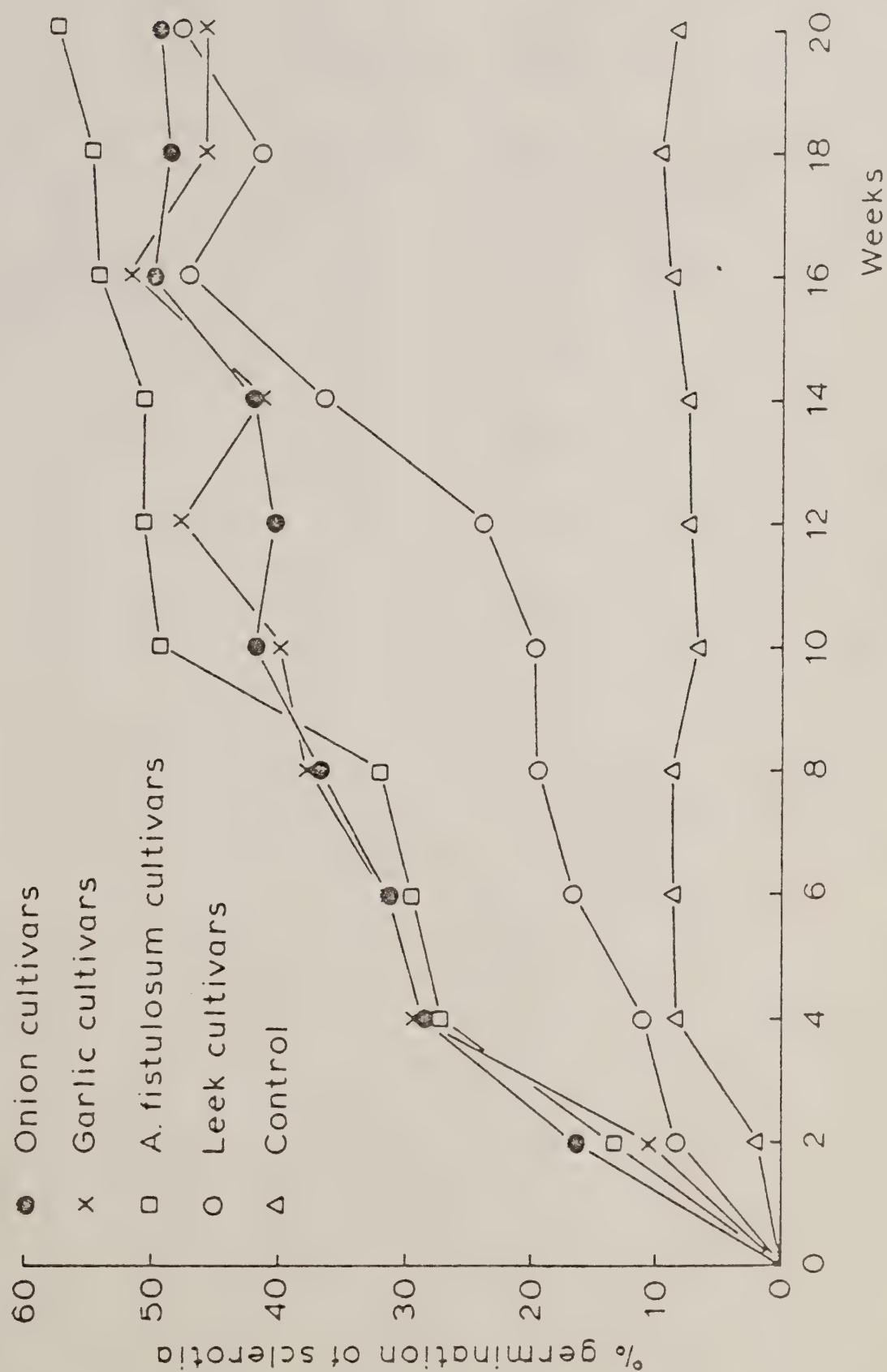


Table 6

End of season analyses of onion and leek cultivars

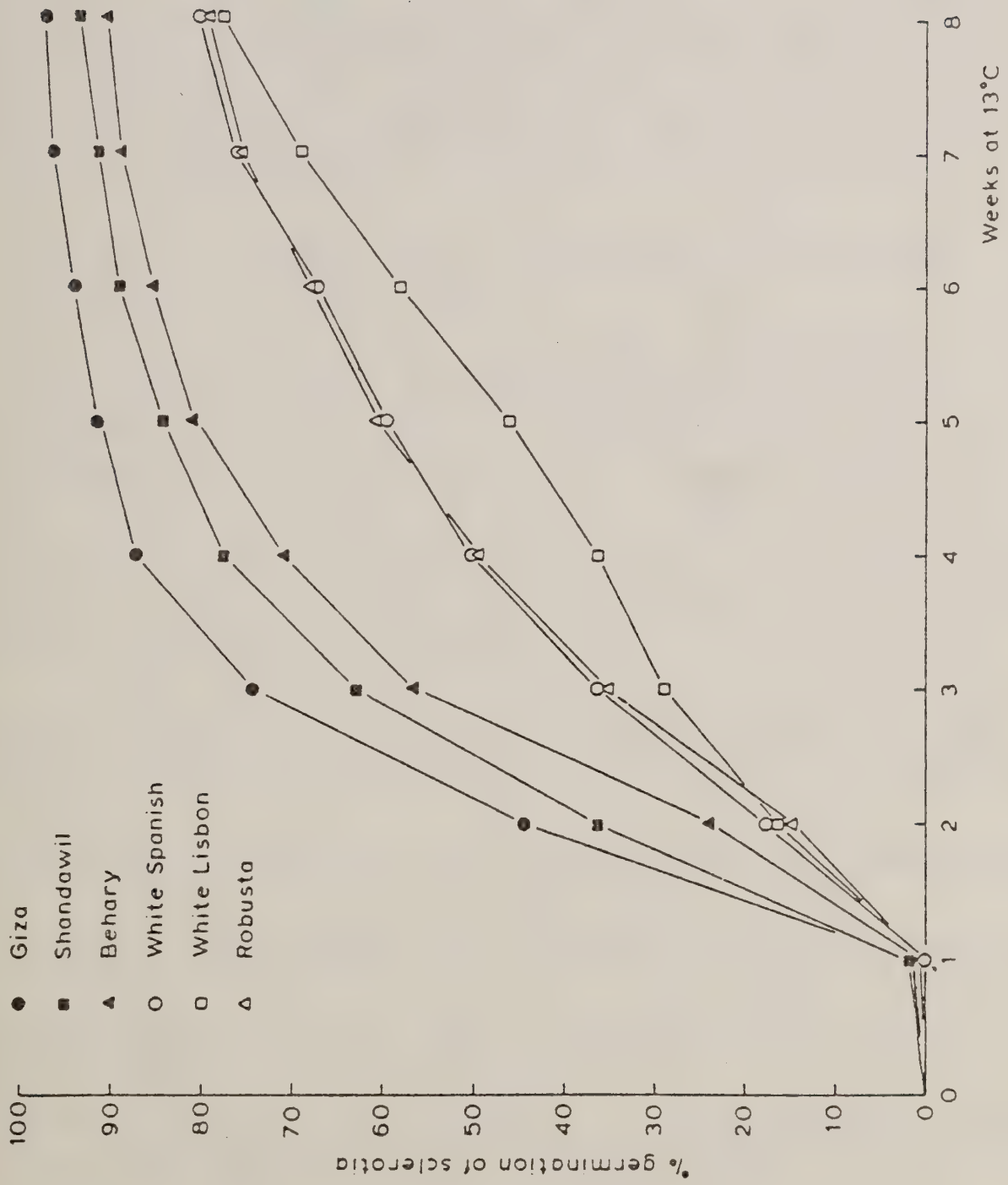
	Onion	Leek
% dry matter	10.6	22.5
% methyl in volatiles	15.3	15.6
Pyruvate μ moles g^{-1} dry weight	75.7	24.8
Pyruvate μ moles g^{-1} fresh weight	7.0	8.8
Thiopropional S-oxide μ moles g^{-1} dry weight	24.5	3.2

Table 7

End of season analyses of Egyptian and European onion cultivars.

	% dry matter	Pyruvate μ moles g^{-1}	
		dry wt.	fresh wt.
Improved Giza	13.3	92.6	16.0
Behary	15.5	83.2	14.8
Shandawil	14.2	90.7	17.6
Robusta	10.8	73.5	8.6
White Spanish	12.5	79.5	12.0
White Lisbon	15.9	65.0	13.5

Fig. 3. Soil tube sclerotium germination experiment.



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We compared 64 commercial cultivars of Allium cepa in muck soil under uniform high levels of inoculum of Sclerotium cepivorum in 1976, and observed significant differences among the cultivars in the incidence of white rot (1). Percentage of white rot in the test populations was lowest in the cultivar Ailsa Craig in both field and laboratory trials. Sixteen cultivars were compared in field trials in 1977 and 1978, and the incidence of white rot was again lowest in the cultivar Ailsa Craig in both years (3). Ailsa Craig from seed purchased in 1979 and 1980 did not show significant resistance (4) to white rot, but Ailsa Craig from seed purchased in 1978 was significantly resistant when retested in the 1980 field trial. Moreover, first generation selections from Ailsa Craig showed the highest levels of resistance when evaluated in the 1980 field trial.

Lines based on recurrent selection from pooled selected individual plant self progenies of Ailsa Craig continue to show significant levels of field resistance to white rot (Table 1).

Two additional sources of resistance to white rot were tentatively identified in 1982. PI 264650, obtained from the USDA world collection of Allium cepa and first identified by us as being significantly resistant to white rot in 1977 (2), and retested in 1982 along with three selections therefrom developed by Dr. W.H. Gabelman of the University of Wisconsin, appears to possess substantial field resistance to white rot. Two hybrids produced by Takii & Co., Ltd. (No. 80-02: Morstar, and No. 10: Eskimo) also looked promising in the field in 1982. (These hybrids were, by mere chance, used as guard rows for field trials in 1982 and thus their performance could not be subjected to statistical analysis. They are being field tested at two sites in formal field trials in 1983.)

We conclude that significant resistance to white rot occurs in commercial onion cultivars, that its level varies in different seed lots of the open-pollinated cultivar Ailsa Craig, and that resistance can be increased significantly via selection.

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Table 1.
Summary of Cloverdale Trial Evaluating Onion Breeding Lines for Resistance to White Rot.

Line Identification	Source ¹	White Rot (%)	Emergence (%)	Bulb Wt. (g) ³	Yield/Plot (kg) ³
PI264650-M 1490-81	WIS-USDA	0.3 a ²	40.0	80 b ²	1.2 gh ²
8KRSL-01	SFU	2.1 ab	36.9	87 b	1.8 efgh
PI264650	USDA	2.1 ab	24.9	94 b	1.0 h
PI264650-M 1492-81	WIS-USDA	4.8 ab	43.2	93 b	1.7 efgh
PI264650-M 1488-81	WIS-USDA	3.4 abc	62.4 abcde	80 b	3.0 bcdef
8KR/4KR-01	SFU	4.6 abc	32.5	100 b	1.7 efgh
SFU-S1-01	SFU	4.1 abc	48.3	92 b	3.2 bcde
4KR0P-01	SFU	5.8 abc	28.2	125 a	2.2 defgh
5KR0P-01	SFU	9.3 abcd	51.8	84 b	2.7 cdefg
AC76P-01	SFU	8.7 abcd	60.6 abcdef	92 b	3.0 bcdef
12KROP-01	SFU	9.2 bcd	54.9 bcdefg	91 b	2.8 bcdefg
W420A4	WIS	10.3 bcde	74.6 abc	78 b	3.8 bcd
PLM#1 1560-81	WIS	14.9 bcdef	51.9 cdefg	88 b	1.4 fgh
(RBM101)MRB/A	WIS	17.9 cdef	82.9 a	97 b	6.0 a
W420A3:W419B4	WIS	21.9 defg	77.5 abc	91 b	4.2 bc
Autumn Spice	COMM	24.0 defg	58.1 abcdef	110 ab	3.4 bcde
Ailsa Craig	COMM	24.8 defg	57.5 abcdefg	127 a	4.7 b
W419A4	WIS	26.1 defg	68.5 abcd	92 b	3.3 bcde
W427A4	WIS	28.7 defg	74.0 abc	86 b	3.4 bcde
W433A2 1515-81	WIS	29.5 defg	79.6 ab	92 b	4.4 bc
75 SYN A1	WIS	30.5 defg	66.5 abcd	108 ab	3.7 bcd
(W404M76SYN)A1	WIS	34.2 fg	65.6 abcd	79 b	3.0 bcdef
W424A4	WIS	35.1 fg	68.5 abcd	98 b	3.2 bcde
W52A4	WIS	43.0 g	79.3 ab	80 b	2.1 defgh
(OM175#11)A1	WIS	44.7 g	79.9 ab	92 b	2.8 cdefg

¹ Source of seed: WIS = University of Wisconsin, W.H. Gabelman; USDA = world germplasm collection of *Allium cepa*, Beltsville, Maryland; SFU = breeding lines developed at Simon Fraser University; COMM = commercial seed companies.

² Means within a column followed by the same letter do not differ significantly (P=0.05) according to SNK test.

³ Values not adjusted for differences in plant populations

Introduction to Session VII: Biological Control

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I'll begin by paraphrasing Jim Cook (4) only slightly in saying that biological control is the use of antagonists to i) manage incompatibility between hosts and pathogens, ii) to protect host plant surfaces from pathogens, and iii) to reduce inoculum of pathogens.

The Allium spp. - Sclerotium cepivorum disease interaction offers a number of attractive possibilities for biological control, particularly as regards reduction of inoculum and protection of host plant surfaces.

At first glance, this disease interaction appears to be a simple case of a susceptible host population eliciting germination of sclerotia of the pathogen, S. cepivorum. Infection of the host follows, new sclerotia are formed on the host, and a proportion of these are dislodged to infest the soil as long-lived dormant propagules, potentially for many years.

A closer look reveals that the white rot disease interaction entails some less obvious complexities. John Coley-Smith and his coworkers (1, 2, 3, 5, 7) have provided evidence which suggests that soil or mycosphere bacteria are a component of the germination response phenomenon via their metabolism of propyl cysteinyl sulfoxide and related components of Allium root exudates to release alkyl and allyl sulfide and disulfide metabolites, these metabolites being potent germination stimulants. Herein lies great potential for biological control.

Can soil sclerotial populations be induced to germinate artificially via the use of germination stimulants? Several researchers, and notably Peter Merriman and his coworkers, have shown that artificial germination stimulants can be used to cause sclerotial germination under mineral soil field conditions, resulting in reduction of both inoculum and disease (6, 8). This approach is presumably biological control as there is strong evidence that germinating sclerotia decay quickly in field soil in the absence of a suitable host.

My colleague Raj Utkhede and I obtained about 50% reduction in sclerotia and 70% reduction in incidence of white rot in a 1981 field trial in muck soil by incorporation of onion oil one month prior to seeding (10). In 1982 we applied onion oil as a furrow drench at seeding and observed no significant effect on the incidence of white rot on muck soil.

A positive effect one year, no effect the next. Why? Different environmental conditions? Different time of application? Perhaps others here have experienced similar frustrations. There's a lot that we don't know about this area, but it appears certain that the potential exists for reduction of inoculum via the use of artificial germination stimulants. John Coley-Smith will present information this afternoon on the response of sclerotia to onion oil applied at different times of the year. His presentation should give new insight into this area, and I expect that some of the rest of us may also have useful information to contribute to this subject.

Sclerotial dormancy and longevity are major factors in the white rot disease interaction. I believe that when we more fully understand these phenomena we will find this disease relatively easy to control biologically.

What don't we know that we need to know? We need to know more about what imposes dormancy. Is it general soil mycostasis or is it a specific mycosphere microflora (5)?

Raj Utkhede and I obtained a number of isolates of S. cepivorum, presumably as pure cultures, from Australia, New Zealand, England, The Netherlands and Canada. Many of these cultures were provided by individuals present here today. We recovered Bacillus subtilis from sclerotia of the majority of these isolates (11). The circumstances of these recoveries suggests to us that there is much more than a casual relationship between B. subtilis and sclerotia of S. cepivorum. The occurrence and function of a specific mycoflora for sclerotia of S. cepivorum has yet to be definitively shown, but given such a possibility, could such a mycoflora be manipulated to advantage?

We need to know what factors affect the longevity of sclerotia in field soil. I'll report evidence this afternoon that shows that sclerotia are comparatively short-lived in some situations. Can we identify the specific factors responsible for rapid decay of sclerotia and use these to advantage?

We need to know what conditions predispose sclerotia to attack by hyperparasites. Can hyperparasites attack fully dormant sclerotia? What environmental conditions are most conducive to activity of specific mycoparasites?

John Coley-Smith, Peter Adams and Tawfik El-Moiety will be providing information this afternoon about Teratosperma, Sporidesmium, Trichoderma and other hyperparasites. Hopefully, others here will be able to contribute information on this subject as well.

The bulb onion gives us at least two beautiful opportunities for application of hyperparasites. One is in the field curing stage. Here we have infected onions with concentrated sclerotia, easily accessible for inoculation during the bulb lifting, curing or topping operations at minimal cost. The second is in storage, which represents an even more environmentally controlled situation. It would be a somewhat radical approach, but sclerotia on infected onions could be inoculated in or from storage and returned to the field after culling.

Can we achieve biological control via protection of host surfaces? Raj Utkhede and I reported success in this regard by onion seed bacterization with B. subtilis (9). The year after that report we had similar positive results, but seed bacterization failed in the next two years. I'll give details of our successes and failures in this regard this afternoon. Prudence Sommerville will report results of her evaluations of seed bacterization for control of white rot of garlic and onion in California. In talking with Prudence earlier today I gather that both of us have had mixed success. At present it seems that this is another area where we presently have more questions than answers, but the potential is there.

Let's think of the biology of the Sclerotium cepivorum - soil microflora - Allium spp. interactions as a giant jig-saw puzzle. Right now it seems that we have most of the pieces identified on the table but we haven't yet put enough of them together to obtain a very clear picture of the biology of the interaction. When we do, I believe that we'll find this disease quite manageable biologically. We have the opportunity to clarify the picture a bit this afternoon.

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Immunofluorescent analysis of rhizosphere populations of Bacillus subtilis in relation to use of seed bacterization for control of onion white rot.

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A fluorescent antibody technique was used to assess the populations of vegetative and spore cells of Bacillus subtilis in onion root rhizospheres of plants grown from bacterized and non-bacterized seeds. The population survey was conducted over several weeks, after planting the seeds in sterilized and non-sterilized muck soils under greenhouse conditions, and muck soil in the field. Rhizosphere populations of B. subtilis were significantly higher for plants grown from bacterized seeds than for those grown from non-bacterized seeds during germination and early stages of plant growth under greenhouse conditions, and persisted longer in sterilized than in non-sterilized soil. In the field trial only one sample period showed a significantly greater population of B. subtilis associated with seed bacterization. Antisera did not distinguish different isolates of B. subtilis and the effects of bacterization with the B2 isolate were soon obscured by B. subtilis presumably indigenous to the soil in which the plants were grown. Resident populations of B. subtilis in the rhizospheres of plants were generally comparable to those occurring in non-rhizosphere soil. We conclude that B. subtilis was not preferentially associated with onion roots, nor was its occurrence in the rhizosphere enhanced in the long term by seed bacterization under the conditions of this study.

Bacteria as Biological Control Agents for Sclerotium cepivorum

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Biological Control Session

Bacteria isolated from the surface of roots of Allium species were screened for their ability to inhibit the mycelial growth of S. cepivorum on potato dextrose, King's B and nutrient agars. Bacteria creating zones of inhibition greater than 1.5 cm were retained, marked with antibiotic resistance markers (100 ug/ml rifampicin and 100 ug/ml naladixic acid), tested for their ability to colonize roots and produce an 'anti-fungal agent.'

Bacteria were applied to seed or cloves using two carriers:

- 1) Bacteria suspended in 1.5% methyl cellulose (in .1M $MgSO_4$) were sprayed on garlic cloves that were then dusted with talcum powder. Onion seed was suspended in the methyl cellulose, talcum powder added and manually stirred to separate the seed.
- 2) A 0.85% saline solution containing 3.0% kappa carrageenan (Colloid Division FMC Corporation) was prepared. Onion seed was immersed in the solution then dropped individually into a cold solution containing 0.3 M KCl and 0.01 M $CaCl_2$ in equal amounts. The carrageenan enclosing the seed formed a gel on contact with the solution. Upon exposure to air, the gel dehydrated rapidly leaving a thin film enclosing the seed. Bacteria survived this dry period and grew when the gel rehydrated in the presence of moisture.

This method was only used on onion seed. A gel in excess of 3.1% carrageenan inhibited germination.

The ability of bacterial isolates to colonize roots once applied to seeds or cloves was tested, in the laboratory in small, plastic cups. The cups were placed in holes drilled in plexiglass template supported over a crisper containing water to a level 1 1/2 inches below the base of the cups. Roots emerged through holes punched in the bottom of each cup. At frequent intervals roots were removed, macerated in normal saline and the suspension was spread over an antibiotic-amended King's B medium. Bacterial colonies were counted 3 days later and colonization counts were recorded as cfu/g dry weight roots (Table 1).

Onion seed coated by methyl cellulose and talcum powder were planted at Tullake, California. Following emergence of plants and at regular intervals thereafter, plants were dug and roots and 1 cm root tip segments were removed and placed immediately into small, stoppered vials containing .1M $MgSO_4$. Root and root tip segments were aggitated for several minutes in washing buffer. This was repeated twice and the final washing solution was retained and spread over an antibiotic medium. Colonization was calculated as cfu/cm root, root tip (Table 2).

Under laboratory condition, Bacillus species generally maintained a higher population on roots originating from carrageenan-coated seed, than did fluorescent Pseudomonas species. The same results were observed under field conditions 10 weeks after planting. Prior to 10 weeks the Pseudomonas species were recovered in a higher concentrations than the applied Bacillus

-3-

species. Early in the season, the populations maintained or the root tips were lower than on the same length of older roots.

'Anti-fungal agents' produced by some of the bacteria antagonistic to S. cepivorum were fungistatic and destroyed when autoclaved. When growing in the presence of certain of the antagonistic bacteria, changes in the colony morphology of S. cepivorum were observed as: thickening and darkening of hyphal cells, also delayed or diminished sclerotia production and also the formation of sclerotia within the media.

The Occurrence of Teratosperma oligocladum and
Laterispora brevirama in Soils in England

D. Parfitt & J.R. Coley-Smith, University of Hull, U.K.

During an investigation designed to survey soils in the U.K. for the presence of mycoparasites of fungal sclerotia two peat soils from a farm in the Southport area of Lancashire with a history of severe Sclerotinia disease were baited with sclerotia of Sclerotinia minor using methods similar to those of Adams & Ayers (1981). Sclerotia of S. minor were produced in Perlite-maizemeal cultures (Esler & Coley-Smith, 1983) and were added to the soils at 2g fresh weight of sclerotia per 100g moist (-500 kPa) soil. The soil-sclerotia mixtures were incubated for 4 wk at 18°C in glass beakers covered with foil to reduce evaporation. The sclerotia were then recovered by wet sieving, transferred to moist filter paper in Petri dishes and kept at 25°C for 4 wk. After this period the sclerotia from both soils were covered with a brown growth of hyphae which proved to be the dematiaceous hyphomycete Teratosperma oligocladum Uecker et al. (1980). The characteristics of the English isolates agree closely with those given by Uecker et al. T. oligocladum has now been found in six soils from Lancashire, one from Humberside and one from Norfolk (Fig.1). The mycoparasite Laterispora brevirama Uecker et al. (1982) has also been found in three of the Lancashire soils which also contain T. oligocladum.

It is becoming apparent that T. oligocladum is widespread in peaty soils in the U.K. but the similar mycoparasite Sporidesmium sclerotivorum Uecker et al. (1978) has not yet been found.

The identity of isolates of T. oligocladum and L. brevirama has been confirmed by Dr. P.M. Kirk of the Commonwealth Mycological Institute.

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Location of
Teratosperma oligocladum
in the U.K.



Biological Control of White Rot of Onion in the Field

by

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Onion white rot is one of the most serious diseases causing high losses for onion growers. Several attempts have been made to eliminate this disease by chemicals or other agricultural means. All these methods can not stop the disease incidence, and the onion production in Egypt dropped from about 354,000 tons in 1963 to only 64,000 in 1982. Studies to control this disease biologically were started in Egypt in 1974. After several lab and greenhouse experiments, we found that certain isolates of Trichoderma harzianum can reduce the disease incidence drastically under certain conditions.

During 1980-81, 1981-82, and 1982-82, 550 Feddan (Feddan = 4200/m²), heavily infested with Sclerotium cepivorum, were treated with T. harzianum (isolate TR-25) growing on Barley grain and gliotoxin fermentation medium (1:0.8

Data obtained showed that the percentage of disease in treated plots was 10-15% compared with 85% disease in untreated plots (data of 80-81). In 1981-82 300 feddans were treated. The general result of efficacy of this treatment was 82% control. During this season, 13 feddans were treated with Iprodione by Rhone Poulenc to compute the effects of biological and chemical control. At harvest time, no difference was noticed between biological and chemical control. In 1982-83, we treated about 230 feddans, and the results obtained by using biological control was about 87% control.

During 1981-82 and 1982-83, the percentage of infection in some treated fields was considerably high. It means that T. harzianum failed to control the

disease in these fields. Evidently, T. harzianum lost its efficacy in controlling the disease under the following conditions:

1. When onions were transplanted after the end of October because temperatures become unfavorable for the antagonist's establishment in the soil.
2. When applying the biocontrol agent in sandy soil. This type of soil is poor in organic matter resulting in poor establishment for the antagonist in soil.
3. Number of irrigations. It was found that more than 2-3 irrigations led to decrease in the efficacy of T. harzianum.
4. Presence of heavy weeds cause reduction in effectivity of T. harzianum.

SECOND INTERNATIONAL WORKSHOP ON ALLIUM WHITE ROT

ABSTRACT - EPIDEMIOLOGY AND DISEASE FORECASTING

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The annual sampling of garlic fields for sclerotial populations of *Sclerotium cepivorum* prior to planting is now an established practice following the development of soil sampling techniques by Dennis Hall, Fred Crowe, and Art Greathead.

The current practice in the Monterey County area is to initiate sampling of garlic fields in July and August, which is 3-4 months prior to planting. This allows adequate time to sample and determine the sclerotial population.

Fields are sampled by obtaining 120 subsamples of soil from each of 4 quadrants and compositing these samples to provide 4 - 1000 gram field samples for analysis. Depth of sampling does not exceed 6 inches. The 4 samples are then assayed to determine the presence or absence of sclerotia and any sclerotia found are then germinated to insure that they represent *S. cepivorum* and not some other organism.

The data obtained from soil sampling is currently being used to predict the best planting/treatment technique involving the in-furrow treatment of Botran. The number of viable *S. cepivorum* sclerotia found determine the subsequent planting procedure as follows:

1. No sclerotia in any sample - plant without treatment.
2. One to five sclerotia in any sample - treat at planting with Botran.
3. More than 5 sclerotia in any sample - do not plant.

Since the advent of pre-plant field sampling, serious problems arising from infestations of this disease have been greatly reduced.

SECONDARY SCLEROTIA PRODUCTION BY SCLEROTIUM CEPIVORUM

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EPIDEMIOLOGY SESSION

THE IMPORTANCE OF SECONDARY SCLEROTIA IN THE EPIDEMIOLOGY OF 'WHITE ROT' IS NOT KNOWN. LABORATORY OBSERVATIONS OF SECONDARY SCLEROTIA FORMATION IN THE PRESENCE OF ALLYL SULFIDE ON FOUR DIFFERENT, HIGHLY PURIFIED AGARS, SUGGESTED THAT S. CEPIVORUM MAY PERPETUATE ITSELF FOR EXTENDED PERIODS OF TIME IN THIS WAY.

SCLEROTIA WERE PLACED IN SMALL PETRI DISHES CONTAINING WATER AGAR AND A MICROBEAKER, PLACED CENTRALLY CONTAINING .02 ML 2% ALLYL SULFIDE. THE PETRI DISHES WERE SEALED AND INCUBATED AT 15°C. THE MICROBEAKERS WERE REMOVED AT VARIOUS TIME INTERVALS. THREE WEEKS AFTER REMOVAL OF THE ALLYL SULFIDE SECONDARY SCLEROTIA FORMATION WAS OBSERVED. THE NUMBER OF SECONDARY SCLEROTIA PRODUCED INCREASED AND THEN REMAINED CONSTANT, 68 DAYS AFTER REMOVAL OF THE STIMULANT. ALL TREATMENTS IN WHICH EXPOSURE TO ALLYL SULFIDE WAS 15 MINUTES OR LONGER YIELDED SOME SECONDARY SCLEROTIA. THE NUMBER OF "SECONDARIES" PRODUCED PER PARENT SCLEROTIUM INCREASED WITH THE TIME OF EXPOSURE AT A DECREASING RATE, AFTER 12 HOURS EXPOSURE.

SCLEROTIA WERE PLACED ON COVERSLEIPS THINLY COATED WITH EITHER PURIFIED, ION, NOBLE OR WATER AGAR. ON EACH MEDIA SCLEROTIA GERMINATED AND PRODUCED SECONDARY SCLEROTIA IF ALLYL SULFIDE WERE ALSO PRESENT. SECONDARY SCLEROTIA WERE ALSO PRODUCED IN THE PRESENCE OF ALLYL SULFIDE, ON A PLAIN COVERSLEIP. THE HIGHEST PRODUCTION OF SECONDARY SCLEROTIA WAS ON PURIFIED AGAR (2 SECONDARY SCL/PARENT GERMINATED).

SECONDARY SCLEROTIA ARE SMALLER BUT OTHERWISE SIMILAR IN APPEARANCE TO PARENT SCLEROTIA. WHEN SURFACE STERILIZED IN 0.5% SODIUM HYPOCHLORITE THEY BECAME BLEACHED AFTER ONE MINUTE AND FAILED TO GERMINATE. IF INTACT, HOWEVER, THEY GERMINATED AND PRODUCED TERTIARY SCLEROTIA IN THE PRESENCE OF ALLYL SULFIDE.

ABSTRACT

Leggett M.E. and J.E. Rahe. Factors affecting the survival of sclerotia of Sclerotium cepivorum Berk. in the Fraser Valley of British Columbia.

Sclerotia of Sclerotium cepivorum buried in muck soil in the Fraser Valley decayed with time. The rate of decay of sclerotia was influenced by local environmental conditions. A mixture of soil with sclerotia increased their survival but there was no difference in the rates of decay of sclerotia in three different soils. The decay was greatest during winter when Fraser Valley fields are often flooded. Sclerotial decay was also affected by pretreatment of the sclerotia. Dried sclerotia decayed significantly ($P < 0.05$) faster than sclerotia which had not been dried, a phenomenon which is apparently due to changes in micro-organisms on the sclerotia. Dried sclerotia which had been incubated in moist soil had fewer bacteria and more fungi than sclerotia which had been incubated in soil without being dried. The increase in fungi on the dried sclerotia was due to a dramatic increase in Trichoderma spp.

The seriousness of white rot of Allium spp., caused by Sclerotium cepivorum Berk., relates substantially to the longevity of the pathogen in soil. Coley Smith [2] reported that sclerotia buried in soil near Hull, U.K. persisted for 10 years with little loss in viability. Crowe [7] recovered viable sclerotia from fields in California where no onions had been grown for 10 to 15 years.

In marked contrast to these reports, we observed substantial decay of sclerotia of S. cepivorum buried in muck soil of the Fraser Valley of British Columbia during less than 1 year [10]. Factors that could underlie differences between our results and those of Coley-Smith and Crowe include biological differences in the sclerotia isolates under test, and soil and/or climatic factors unique to the Fraser Valley.

This paper describes further studies on the rates of decay of untreated and dried sclerotia of S. cepivorum in the Fraser Valley, and evaluates the effect of soil and soil type on their survival. Variations in climatic factors in the Fraser Valley over the three years of studies on survival of sclerotia are compared with the average of these factors for the Fraser Valley of B.C. and Hull, U.K. The effects of drying of sclerotia on their associated fungi and bacterial populations are also reported.

Integrated Control of Sclerotium cepivorum

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Introduction. The term integrated control describes a situation where one form of control is combined with at least one other. This is done in order to achieve a level of success which is either better than the two or more forms give individually or because there is some other advantage such as a decreased usage of pesticides.

At the present time integrated methods are not consciously being practised for controlling S. cepivorum and it seems likely that in the foreseeable future the use of fungicides will continue to be the most important method of control. It is perhaps appropriate therefore to consider what the long-term future for these fungicides is likely to be.

The main groups of fungicides which have been or are being used for control of white rot are mercurials, aromatic hydrocarbons e.g. dicloran, benzimidazoles and recently the dicarboximides. Some members of the triazole (e.g. triadimefon) group have also been used (Entwistle & Marian 1983). How likely is it that resistance of S. cepivorum to any of these fungicides will develop? The development of resistance to fungicides depends on a number of factors, important amongst which are the chemical nature of the fungicide, its mode of action, the nature of the pathogen and the nature of the disease it causes. The risk factor associated with the different fungicides used against S. cepivorum varies. Table 1 gives the general risk factor with plant pathogens. With S. cepivorum the risk would probably be relatively low. Differences between isolates have been claimed but fresh field isolates appear quite uniform. The fact that S. cepivorum has no sexual stage must limit its capacity for variation. Even if fungicide resistant strains did arise they could remain spatially isolated for some time with care. In the case of the dicarboximides and triazoles we should also consider the fact that resistant strains of other fungi are frequently less fit than sensitive strains and because of this field resistance has often failed to become a serious problem.

The main question to be asked about fungicides is whether they are sufficiently effective against white rot. In the U.K. the dicarboximides, particularly iprodione, do seem to be effective with salad onions but are much less so with bulb onions. There does therefore seem to be some point in looking for other methods which could be integrated with the use of fungicides in order to improve the degree of control in the bulb crop. The remainder of this contribution will be devoted to a consideration of possible ways of doing this.

Reduction of propagule numbers.

S. cepivorum can survive in field soil for many years. The latest results which we have from a long term replicated field trial are shown in table 2. There has been little decrease in numbers of sclerotia after 15 yrs burial.

Primary infections arise from sclerotia and it is highly likely that in the bulb crop most infections are primary. In the salad crop the situation is different with most infections being secondary and resulting from plant to plant spread (Entwistle & Munasinghe 1978). It follows that reductions in sclerotial number should be of most significance in the bulb crop. What we need to know is by how much sclerotial populations will need to be reduced. This clearly depends upon the relationship between sclerotial density and infection. Population densities can be as high as 1000 sclerotia per kg soil and from the data of Crowe et al. (1980) it can be calculated that the density would have to be reduced to around 10 sclerotia per kg or less in order to effect a significant reduction in white rot. In other words whatever method is used it has to be capable of reducing high sclerotial densities to around 1% of their untreated levels. At lower sclerotial densities, which are common, this would not of course be the case.

The question which has to be asked is whether any methods now available are capable of effecting such reductions. There are several methods which could be considered, the most obvious ones being the use of germination stimulants and the use of sclerotial mycoparasites.

(a) Germination stimulants.

There have been several investigations of the effect of germination stimulants (Entwistle et al. 1982; Merriman et al. 1980; Utkhede & Rahe, 1982) and we are now in a position to draw certain conclusions. Firstly it is clear that some reduction in sclerotium densities can undoubtedly be achieved. It seems to me that efforts should now be directed towards maximising this effect. There seems little point in using artificial or natural onion oils since diallyl disulphide is just as effective, perhaps more so. Because of the likely production costs the use of non-volatile sulphide precursors cannot be envisaged. Unfortunately results even with diallyl disulphide have been variable and the reasons for this variability are not known. My most recent work does indicate seasonal differences in response of sclerotia with the poorest results arising from midsummer treatments, but there could also be variation in the response of different field populations of sclerotia. In my most recent experiments reductions of up to 95% have been achieved but it is unlikely that such levels would be realised in practice.

(b) Sclerotial mycoparasites.

It has been pointed out (Papavizas & Lumsden, 1980) that mycoparasitism has not been very well explored and is poorly understood in relation to survival of fungal propagules in soil. This is certainly true in the case of S. cepivorum. Sclerotia of this fungus are obviously very resistant to microbial attack but there are a number of organisms which are capable of invading them. The most important of these are probably Coniothyrium minitans and the more recently discovered Sporidesmium sclerotivorum (Uecker et al. 1978) and Teratosperma oligocladum (Uecker et al. 1980). Coniothyrium and Sporidesmium have been shown to give some control of Sclerotinia species and Sporidesmium has been held responsible for the natural decline of sclerotia of both Sclerotinia and S. cepivorum in a number of field soils (Adams & Ayers, 1981). Is it possible to exploit

the activities of these mycoparasites? Coniothyrium is widespread in soils throughout the world but although it has been shown to be capable of invading sclerotia of S. cepivorum little has been done to investigate its possible effects on field populations. Sporidesmium has not been found outside North America and appears to have a more restricted distribution than Coniothyrium. Teratosperma has now been found in the U.K. but although not uncommon it does seem to be restricted to certain types of soils. It is interesting that my experiments on long term survival of S. cepivorum sclerotia have all been done in soils which we now know to be free from Coniothyrium, Sporidesmium and Teratosperma. Could this be a reason for the apparent discrepancy between the results of Adams & Ayers (1981), who found Sporidesmium to be associated with a decline in numbers of sclerotia of S. cepivorum, and my own where the sclerotia have so far survived for 15 yrs? It does seem to be worth attempting to introduce Sporidesmium or Teratosperma into soils which do not already contain them. The introduction of organisms into an alien environment is often fraught with difficulties but Adams & Ayers (1981) appear to have done it successfully. If a mycoparasite can be successfully introduced and maintained then in the short term there could be quite violent oscillations in the levels of the plant pathogen and its mycoparasite (Deacon, 1983). In the long term a steady state condition would probably be achieved with plant pathogen and mycoparasite in a rough state of balance but with the plant pathogen, i.e. S. cepivorum, at a lower level than it would be without its mycoparasite (Alexander, 1981).

What other possibilities exist for reducing sclerotial populations:-

(c) Soil partial sterilants.

We have already heard something about the use of metham-sodium for controlling white rot in bunching onions. This is a material with a fumigant action the activity of which results from its decomposition to methyl isothiocyanate. Allyl isothiocyanate is a reasonably effective

germination stimulant for S. cepivorum sclerotia (Coley-Smith et al. 1981) although it is not a constituent of Allium spp. I have tested a range of concentrations of methyl isothiocyanate and have found no stimulatory activity. This compound almost certainly works therefore by killing sclerotia and other soil partial sterilants would probably work in the same way.

(d) Solar heating of soils.

Considerable progress has been made with this method with some soil-borne plant pathogens (Katan, 1981). It usually involves mulching the soil with transparent polyethylene sheets. In countries like Egypt and Australia the method obviously has potential for control of white rot but in northern Europe it is unlikely to be effective although White & Buczacki (1979) did obtain some control of clubroot disease of Brassicas with it in England. Their experiments were done in the hot summer of 1975. In more normal summers there is unlikely to be sufficient sunshine.

All of the methods discussed could easily be integrated with the use of fungicides. A major function of the fungicides would be to reduce or delay the replenishment of sclerotial populations.

Reduction of propagule germination

If permanent reductions in sclerotial populations cannot be achieved then primary infections could be affected by adopting measures designed to prevent the sclerotia from germinating. At present we have little idea of whether this is possible but what is clear is that fungicides used at rates designed to control white rot do not work in this manner. Iprodione (Entwistle & Munasinghe, 1980) has been shown to have little effect on germination of sclerotia and in my recent experiments I have found that this is also true of vinclozolin and a number of benzimidazoles. There are however a number of methods which might act by reducing sclerotial germination.

(a) Biological control.

There have been a number of attempts to control white rot biologically

and some of these have been at least partially successful. Organisms involved are Coniothyrium minitans (Ahmed & Tribe, 1977), Penicillium nigricans (Ghaffar, 1969), Bacillus subtilis (Utkhede & Rahe, 1980) and Trichoderma harzianum (Abd-El Moity & Shatla, 1981; Papavizas et al., 1982). Most of these methods have not given a very high degree of control but one difficulty is that we have no idea how any of the systems work. Methods involving biological procedures often require a very detailed knowledge of the mechanisms involved before they can be made to function adequately. It is possible that some of the antagonists work by suppressing sclerotial germination. Sclerotia in soil are normally held in check by mycostasis and are stimulated to germinate in the presence of Allium spp. If the level of microbial activity in soils could be raised sufficiently then even Allium spp. might not overcome the inhibition. If antagonists are found to work in this way then it should be possible to investigate ways of improving their germination suppressive activities. This is probably a better approach than adding antagonists to soil and looking for reductions in disease. To be reasonably effective potential antagonists would probably have to work well at fairly low temperatures and would have to be fairly resistant to the commonly used fungicides. The soil tube method of measuring sclerotial germination (Esler & Coley-Smith, 1983) could easily be adapted for investigations of this kind.

(b) Cultural practices.

Effects on white rot disease of cultural practices, such as fertiliser regimes and irrigation, have been claimed (Sirry et al. 1974, a,b) but have not been fully explored and the ways in which they work are not known. In order to discover whether any such practices might affect sclerotial germination a much more detailed knowledge of sclerotial germination in the field must be obtained. It is becoming clear that in the U.K. spring sown plants are most at risk from germinating sclerotia early in the growing season. In summer there is often a fall in sclerotial germination and when suitable

conditions develop again in late summer and early autumn the crop is too mature to be in much danger. If ways could be found of reducing the risk in the early season then some success might be achieved. One obvious way of reducing the risk is to avoid damage to plants. In Egypt it is common practice to use transplants rather than direct seeding. This inevitably results in damaged roots which release much larger quantities of stimulants than undamaged ones. The same is true of plants grown in multiseeded peat based blocks, a method which is on the increase in the U.K. These blocks are at first joined together and are separated at planting time. This results in considerable root damage and the response of sclerotia under such block raised plants is much more rapid than under direct seeded ones. This year we are doing a very detailed trial in which the behaviour of sclerotia under a variety of planting systems is being investigated. Although results from this are not yet available I am sure that it is the sort of investigation which needs to be done if we are to have any chance of selecting cultural systems which might act by reducing sclerotial germination.

(c) Cultivars.

Finally let me consider the effect of different onion cultivars. From what has been published recently (Utkhede et al. 1982) it does appear that small differences in the susceptibility of different cultivars do exist but they are only detectable in large scale trials and the basis for the differences has not yet been conclusively established. What evidence there is suggests that the differences are established before infection (Rahe, 1981) and it is assumed therefore at the sclerotial stimulation level. To look for small differences in stimulatory activity amongst a large number of cultivars would be a vast undertaking. Since the sclerotial stimulants are derived from flavour and odour precursors however I have decided to investigate possible differences in stimulatory activity by examining flavour and odour levels in a large number of cultivars. If cultivars with high and low levels can be identified then the effect of these on sclerotial germination could be

tested. There are many pitfalls in this type of work, particularly when using cultivars from different parts of the world which might not be well adapted to the local climate. If however cultivars with a low capacity to stimulate germination of sclerotia could be selected then obviously the use of such cultivars in an integrated control system is a very attractive idea. Resistance, even if based on a disease escape mechanism, would be the easiest of all features to integrate with the use of fungicides.

Conclusion

Although integrated procedures for controlling white rot disease are not being practised a number of possibilities exist of doing so. The satisfactory development of such systems would require more thorough basic research than is being undertaken by most workers at present.

Acknowledgement

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Table 1

Fungicide or group	Mechanism of action	Whether systemic	Occurrence of resistant strains		Risk factor in disease control
			<u>in vitro</u>	<u>in vivo</u>	
Hg compounds	Multisite inhibitors	-	+	+	Low
Aromatic hydrocarbons	Not known	-*	+	+	High
Benzimidazoles	Interfere with mitosis	+	+	+	High
Dicarboximides	Not known	-*	+	+	Moderate to high
Triazoles	Inhibit sterol biosynthesis	+	+	-	Low

* A few have some systemic activity e.g. Chloroneb and procymidone
From Dekker (1981) and Kaars Sijpesteijn (1982).

Table 2Survival of pure culture sclerotia of S. cepivorum in the field

Depth of burial (cm)	Time of recovery (years)	Percentage recovery of sclerotia	Percentage viability of recovered sclerotia
0	10	60*	90
7.5	10	96	87
15.0	10	100	90
22.5	10	100	62
0	15	42*	90
7.5	15	90	98
15.0	15	96	97
22.5	15	88	98

* Nylon bags split, some sclerotia may have been lost.

Response of Sclerotia to Artificial Stimulants at
Different Times of the Year

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Introduction.

A number of studies have been made of the effect of artificial germination stimulants on sclerotia of S. cepivorum (Entwistle et al. 1982; Merriman et al. 1980; Utkhede & Rahe, 1982). Some variation in response of sclerotia has been detected and in view of this an investigation of possible causes of this variation has been started.

Methods.

Artificial onion flavouring (C7713) and liquid onion concentrate (FD 6039) were supplied by Bush Boake Allen, London. Wetting agent was Agral (containing 90% alkyl phenol ethylene oxide condensate) supplied by ICI, Plant Protection Division. Laboratory soil-tube experiments (Coley-Smith, 1960) were done with S. cepivorum isolate J11 (ATCC 11793) and field experiments with batches of sclerotia collected from a naturally infested field plot. For field experiments nylon bags each containing 50 sclerotia and a small quantity of sand were buried at 10 cm depth in lengths of Netlon 2.5 ml of stimulant was injected on to each bag at appropriate times.

Results.

Field Trial 1. 1980

Results for this trial are given in Table 1. Best results were obtained with combinations which included an April treatment. Treatment in July was less effective than in April. No conclusions can be drawn regarding the effectiveness of the October treatment since sclerotia were still germinating at recovery time and the final survival figures could have been much lower.

Field Trial 2. 1981-2

Results for this trial are given in Table 2. The poorest results were obtained with the July treatment. Surprisingly good results were obtained with a January application and although the response of sclerotia to this was slow, the final effect on sclerotial recovery was considerable (Fig.1). Response of

sclerotia to injections at other times of the year was more rapid.

Laboratory Experiment on Effect of Constituents of Onion Flavouring C7713

The effects of the constituents of C7713 are shown in Table 3. It is clear that diallyl disulphide is the most active of the constituents but allyl isothiocyanate is also moderately effective. This is surprising because it is not a constituent of Allium species but is found in certain crucifers.

Laboratory Experiment on the Effect of Onion Flavourings and Diallyl disulphide

The results of this experiment are shown in Table 4. They show that diallyl disulphide is just as effective as artificial onion flavouring and liquid onion concentrate. The addition of Agral to diallyl disulphide made the latter easier to handle and appeared to have little effect on germination of sclerotia except at the highest concentration.

Conclusions.

It is apparent that seasonal variation in the response of sclerotia to artificial stimulants does occur. Midsummer treatments in the UK are less effective than treatments made at other times of the year. Diallyl disulphide was as active as any of the other materials tested and can be mixed with a wetting agent such as Agral without appreciable loss of activity. Other possible causes of variation in response of sclerotia to artificial stimulants are being tested.

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Table 1

Field Trial 1, 1980. Effect of Concentration of Onion Flavouring C7713 and Time of Treatment on Survival of Sclerotia. Randomised block design with 4 replicates.

Treatment	% survival of sclerotia
(A)	
Nil	87.7
April	26.7
July	40.2
October	72.0
April + July	22.0
April + October	23.8
July + October	32.7
April + July + October	22.3
(B)	
1% Onion flavouring	38.1
5% Onion flavouring	32.5
10% flavouring	32.1

(A) Summed over C7713 concentrations

(B) Summed over times of treatment.

Table 2

Field Trial 2, 1981-2. Effect of Onion Flavouring C7713 (5%) Applied at Different Times of the Year on Survival of Sclerotia. Randomised block design with 6 replicates.

Time of application	% survival of sclerotia
April 1981	4
July 1981	28
October 1981	2
January 1982	2
Control (untreated)	83

Response of sclerotia following a January 1982
application of artificial onion flavouring C7713 (5%)

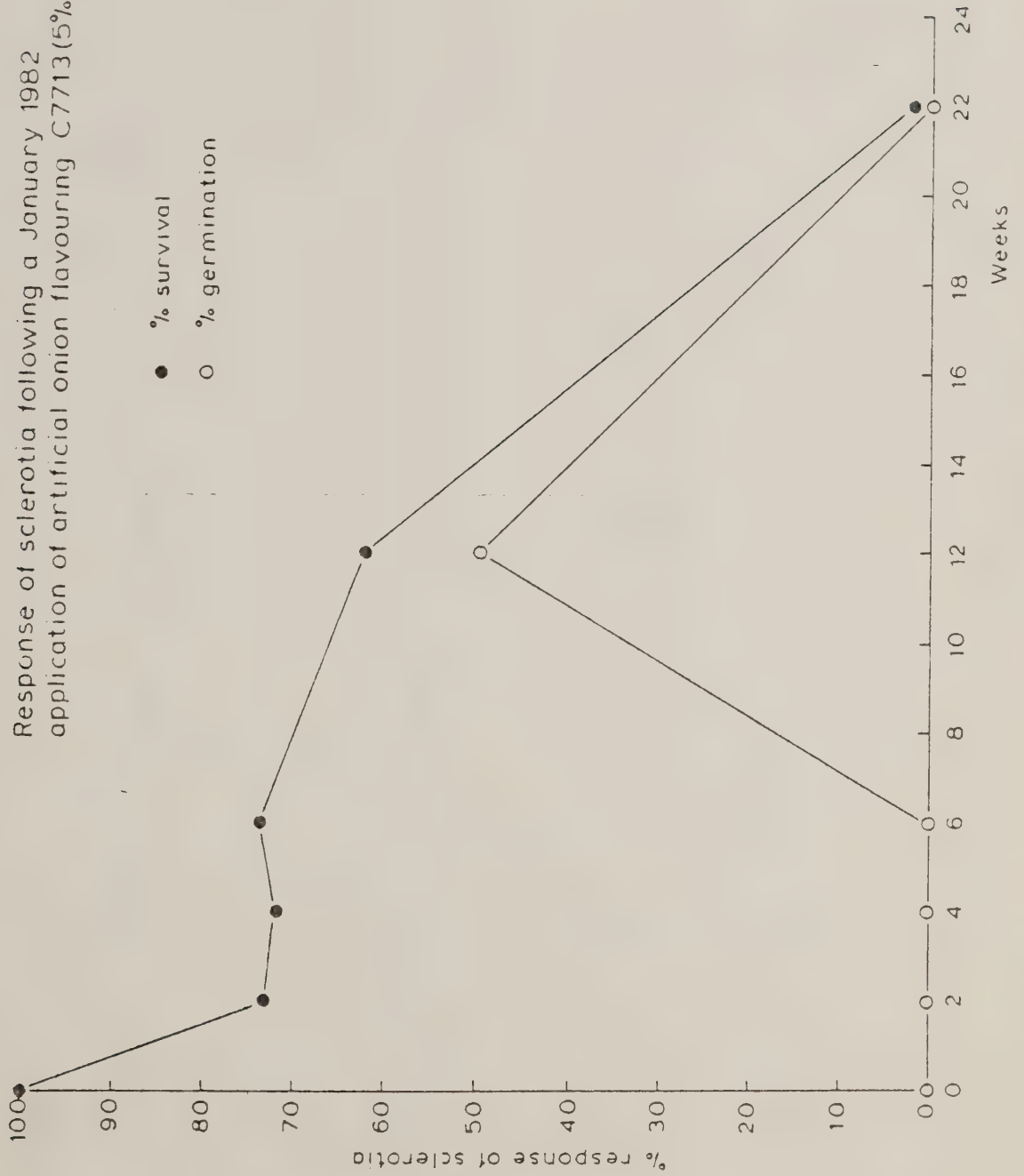


Table 3

Laboratory Experiment on Effect of Constituents of Onion Flavouring C7713 on Germination of sclerotia (%)

Concentration of compound in water	Diallyl disulphide	Di-iso-propyl disulphide	Di-n-propyl disulphide*	Allyl isothiocyanate	Allyl alcohol
0.1 mg l ⁻¹	16	7	3	7	0
1.0 mg l ⁻¹	48	3	4	18	4
10.0 mg l ⁻¹	62	3	22	66	0
100.0 mg l ⁻¹	68	11	50	61	2

Distilled water control gave 3% germination

* Not a constituent of C7713 but included as a control for the Di-iso-propyl disulphide

Table 4

Laboratory Experiment on Effect of Onion Flavouring C7713, Liquid Concentrate FD6039 and Diallyl Disulphide on Germination of Sclerotia (%).

Concentration (%)	C7713	FD6039	Diallyl disulphide	Diallyl disulphide + Agral*
4.86	64.0	97.5	89.5	27.5
1.62	91.5	99.5	89.0	93.5
0.54	100.0	100.0	99.5	100.0
0.18	99.5	100.0	100.0	99.5
0.06	99.0	100.0	100.0	100.0
0.02	100.0	100.0	100.0	99.5

Distilled water control gave 23.5% germination.

* Concentrations of Agral were the same as those of diallyl disulphide.

Effects of diallyl disulphide and Iprodione on sclerotia of *Sclerotium cepivorum* and incidence of white rot in dry bulb onions

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Our most recent experiments on white rot have been concerned with disease control at a site near Colac, Western Victoria. This district has significance because, in the decade beginning 1940 it was Australia's major centre for onion production. Its importance gradually declined following the establishment and spread of *Sclerotium cepivorum*, and currently there are estimated to be some 4,500ha of the Colac district contaminated by the pathogen. So far all attempts to control white rot in these soils have been unsuccessful.

The experiments reported here tested the effects of diallyl disulphide and Iprodione in soil where the population of sclerotia varied between 50 and 150 per kg soil; and where disease prevented the production of dry bulb onions. Iprodione was selected because of its effects against white rot in salad onions (Entwistle and Munasinghe 1980), and diallyl disulphide because it reduced numbers of sclerotia in soil by up to 70% in comparison with controls (Entwistle et al. 1982). Also, tests with artificial onion oil, a formulation containing diallyl disulphide, showed that it reduced incidence of white rot in dry bulb onions at Werribee (Southern Victoria) from 57 to 13% in comparison with controls. However the same treatment applied to Colac soil had no effect on disease. The discrepancy between sites is thought to be due to differences in the residual population of sclerotia after treatment.

For example, after treatment with artificial onion oil, the number of sclerotia per kg soil was reduced from 44 to 13 (70%) at Werribee and from 187 to 87 (53%) at Colac. Apparently the reduction in numbers of sclerotia at Colac was inadequate to influence disease, and this raised the question of the effect of additional treatments on the population of sclerotia and incidence of white rot. This together with the effect of Iprodione on disease were examined at the Colac site from 1980 to 1982.

Artificial onion oil is a synthetic product whose formulation is protected for commercial considerations. In order to achieve more precision in calculating rates of application diallyl disulphide, a component of artificial onion oil which has comparable effects on sclerotia (Entwistle et al. 1982), was selected for further work.

Soil at the experimental site was prepared as a seed bed and kept weed free. Plots (dimensions 2m x 1m) were treated two or three times with diallyl disulphide on 26th August, 1980, 26th May, 1981, and 7th June, 1982. Dates of application for plots receiving the double treatment were 26th August, 1980, and 26th May, 1981. A wetting agent, Teric 17 A3 (I.C.I. Australia), was mixed with diallyl disulphide prior to diluting it in water to a concentration of 0.1%. This was injected in soil at 25cm centres and 10cm depth at a rate equivalent to 400l/ha. The population of sclerotia in soil was assessed 11 weeks after the last treatment with diallyl disulphide and one day before sowing. Thirteen soil cores (dia. 1cm) were sampled from each plot at 50cm centres to a depth of 20cm. The samples from each plot were pooled, mixed and the wet sieving method (Merriman et al. 1980) was used to determine the number of sclerotia in two 100g subsamples of moist soil from each composite sample.

Five rows of onion seed c.v. Pukekohe were sown on 16th July, 1981. Iprodione (50 W.P.) was applied as a seed or seed and stem base spray (S.B.S.) treatment at rates of 100g a.i. per kg seed and 15g a.i. per 2 litres water per 100m row (3kg a.i./ha). The S.B.S. was applied 5 weeks after emergence when plants were at the two leaf stage. Soil, seed and plants in control plots were untreated, the trial design was a randomised block with treatments replicated six times. At monthly intervals during the growing season the plant population in the centre row of plots was counted and expressed as a percentage of the number of plants which emerged. Dying and dead plants were checked at each assessment for symptoms of white rot. At maturity the incidence of white rot, and the individual and total weight of healthy bulbs per plot were determined. The experimental site was not irrigated.

The results showed that, in comparison with the controls, treatment with diallyl disulphide at two or three times reduced numbers of sclerotia by between 30 to 50% (Table 1). There was no difference between diallyl disulphide treatments. The double application of diallyl disulphide dramatically increased plant deaths over all other treatments (including diallyl disulphide x 3) at assessment dates on the 16th October, 13th November and 14th December (Fig. 1). Plant deaths were reduced by Iprodione seed + S.B.S. alone or in combination with diallyl disulphide x 3, when compared with controls. There was no effect of the treatments on final yield, but Iprodione seed + S.B.S. in combination with diallyl disulphide x 3 increased the weight of individual bulbs.

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Table 1

Effects of diallyl disulphide and Iprodione treatments
on numbers of sclerotia, incidence of white rot
and yield of dry bulb onions

Treatments	No. of sclerotia /kg soil (0-20cm)	Overall % plant deaths due to white rot	Yield healthy bulbs (t/ha)	Wt/bulb (g)
Nil	87	53	5.2	61
Ip seed	93	39	4.4	67
Ip seed/SBS	90	34*	5.2	67
DAD(x2)	50**	72*	3.8	64
DAD(x3)	50**	53	5.0	67
Ip seed/DAD(x2)	60**	36	3.0	60
Ip seed/DAD(x3)	54**	40	4.2	58
Ip seed/SBS/DAD(x2)	57*	40	5.8	67
Ip seed/SBS/DAD(x3)	43**	34*	7.0	88*

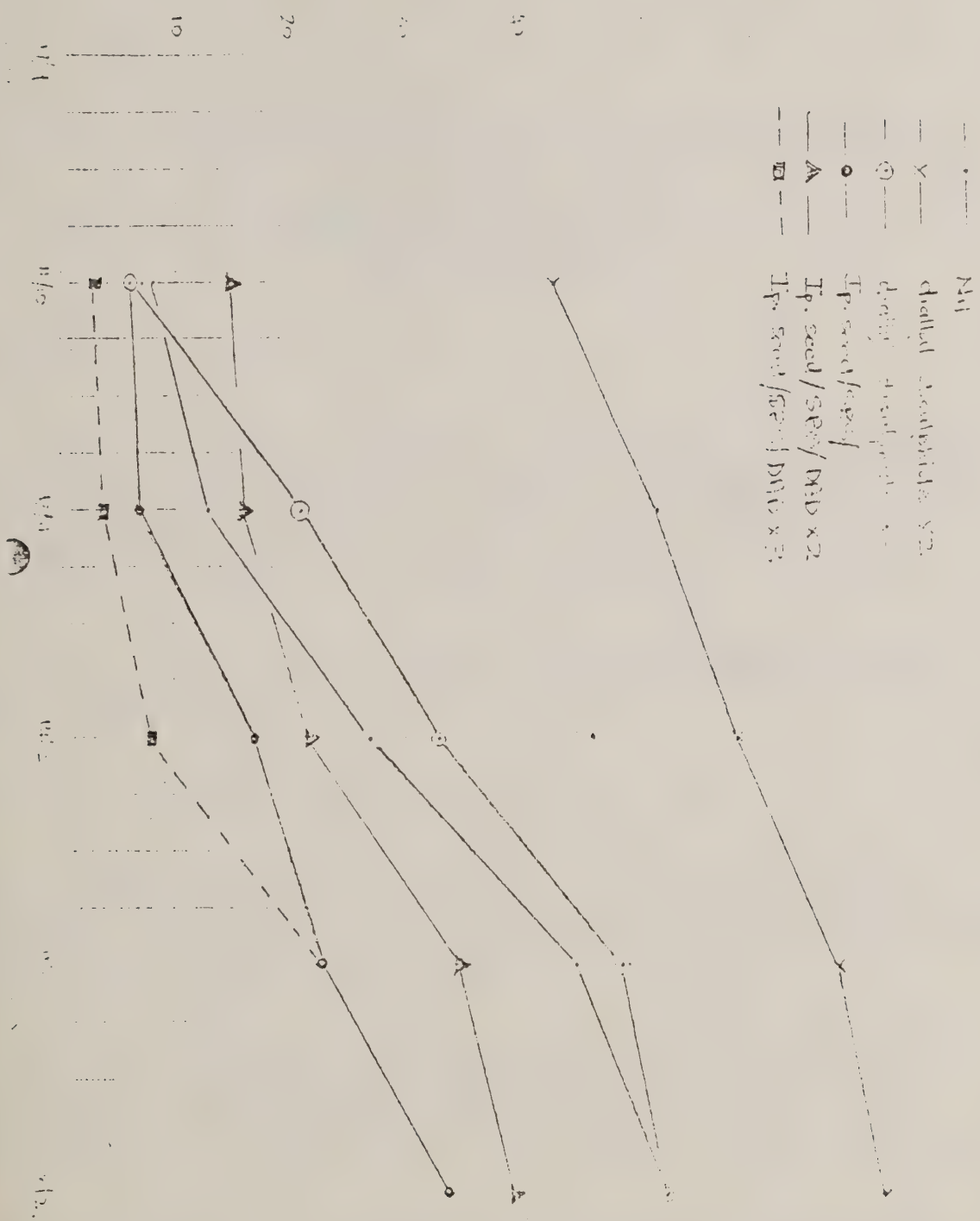
* significantly different from Nil at 0.05

** significantly different from Nil at 0.01

Ip = Iprodione

SBS = stem base spray

DAD = diallyl disulphide



EFFECT OF INOCULUM POTENTIAL ON CONTROL OF WHITE ROT BY FUNGICIDES

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(Integrated Control Session)

Control of white rot disease of garlic and onions by means of in-furrow applications of fungicides has often given erratic results from year to year with the same fungicide applied in the same way. Crowe (unpublished) found in a trial conducted in the Salinas Valley of California that increasing the inoculum potential resulted in an increase in numbers of infected plants regardless of the fungicide used. From these preliminary findings further work on the effect of inoculum potential on chemical control was begun.

A trial was established at Davis, CA, to investigate the effect of different inoculum levels on control of white rot of garlic with the fungicides dicloran (Botran, Tuco Chemical Division of Upjohn) and iprodione (Rovral, Rhone Poulenc, Inc.).

Sclerotia used to infest soil were produced on sterilized wheat seed in glass jars and collected sclerotia stored dry until used.

Appropriate numbers of sclerotia for the different inoculum levels for each plot were determined by weight, mixed with 150 grams of white corn meal which provided enough bulk to spread sclerotia evenly over the surface of pre-formed beds. The sclerotia were then incorporated into the top 10 cm by hand with a "clam" fork. Inoculum was added to approximate 0.5, 0.1, 0.01, 0.005 and 0.0 sclerotia per gram of soil.

After incorporation of sclerotia, furrows approximately 5 cm deep X 10 cm wide were made by hand. Fungicides were applied, by hand-held sprayer, to the bottom and sides of the furrows of the appropriate plot. Dicloran was used at 0.173 gms/30 cm and iprodione at 0.069/30 cm of bed. Sixty garlic cloves of cv. California Early were planted in each plot and covered by hand.

Plots were two meters long and replicated 4 times. Cloves were planted November 17, 1980, plant counts made February 9, 1981, and final disease counts made May 15, 1981.

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Effect of inoculum potential on control of white rot of garlic
by in-furrow applications of fungicides

Treatments	Rate lbs a.i./A	Percent White Rot ¹ Calculated Inoculum - scl./gram				
		0.5	0.1	0.01	0.005	0.0
No fungicide	--	94.2a	95.6a	68.5a	42.9a	6.5
Dicloran	7.5	44.9b	27.9b	3.8b	11.8b	5.0
Iprodione	2	23.9c	10.2c	5.1b	6.6b	0.3

¹ Average of four replications.

The results are given in the Table. There were over 94% infected plants in those plots not treated with a fungicide at the calculated levels of 0.5 and 0.1 sclerotia/gram. The incidence of white rot in the untreated plots at inoculum levels of 0.01 and 0.005 per gram were lower than expected. This suggests that the actual levels of viable sclerotia were lower than those calculated.

The data illustrates that inoculum potential can markedly affect control when fungicides are used as described here. As the inoculum level decreased, control with fungicides improved.

"Avoiding whit rot disease through growing onion from sets."

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Onion growing from sets is practiced in Egypt a long time ago. Because of escaping the white rot disease to some extent, the area of this crop is increasing, while that grown from transplants as winter crop is decreasing because of the high infection of the bulbs by white rot disease. While the average area of onion from sets is increasing from 6257 acres during the period (1963 - 1972) to an average of 11339 acres during the period (1973 - 1982), the average area of winter crop onion devoted for exportation is decreasing from 41220 acres during the period (1963 - 1972) to an average of 21157 acres during the period (1973 - 1982). Some growers use the large size sets in growing their crop, so they usually have a high percent of bolters and doubles, the others grow the small bulbs sorted from the winter crop and in this case the percent of bolters and doubles reaches 90 to 95 percent or more. In such cases, the bulbs produced are not of proper maturity and of inferior quality and are used only for local consumption and in dehydrated onion process.

Through a Dutch - Egyptian project and technical assistance of the Dutch experts a proper size of sets (8-16 mm) is available, and when grown during August or early in September for bulb production a maximum of 5 percent bolters is reached, and a high percent of exportable sound bulbs maturing during December is produced. The doubles are minimized by growing the sets 5 cms apart. To escape the white rot disease to a high extent, an extra early maturing cultivar is needed. Many cultivars and onion strains were tested during the last 4 years. Beth Alpha and Shandaweel.1. cultivars were earlier in maturity than the commercial cultivar Giza 6 M by 3 and 2 weeks respectively.

(2)

On September 16, 1982 fifteen cultivars and strains of onion sets 8 - 16 mm in diameter were grown in a field plot where severe damage with white rot disease had been observed in onion seed crop last season at Tameia state farm at Fayom province the following table shows the infection percentage recorded at time of harvest for each entry.

Entries	Date of Harvest	Total Plants tested	Plants affected by <i>S. cepivorm</i>	
			No.	%
1. Beth Alpha.	Dec. 20, 1982	376	15	4.0
2. Giza 20 (1361)	Feb. 3, 1983	474	273	57.0
3. Behairy (Zefta).	Feb. 3, 1983	480	213	44.4
4. Yellow Bermuda Excell 986	Feb. 3, 1983	116	35	30.2
5. Extra Early Yellow Bermuda	Dec. 20, 1982	465	7	1.5
6. Giza 6 Globe.	Feb. 3, 1983	617	210	34.0
7. Giza 6 Mohassan.	Feb. 3, 1983	331	93	28.1
8. Shandaweel. 1.	Jan. 2, 1983	743	181	24.4
9. Texas Early Yellow Grano	Feb. 3, 1983	202	83	41.1
10. Comp. 16 (2693).	Feb. 3, 1983	1102	411	37.3
11. Comp. 16 (2029).	Feb. 3, 1983	595	198	33.3
12. Giza 6 Mohassan (2963)	Feb. 3, 1983	1130	280	24.8
13. Giza 20 (Nuba seed).	Feb. 3, 1983	915	313	34.2
14. Shandaweel. 1. (S.Tahrear).	Jan. 2, 1983	486	97	19.9
15. Giza 6 M. (Ismaelia).	Feb. 3, 1983	3146	1017	32.3

(3)

Putting in consideration the unusual cold and long winter this season in Egypt that causes much delay in maturity of the bulbs all over the country, the cultivars Extra Early Yellow Bermuda, Beth Alpha, and Shandaweel 1. showed the least percent of infection. More over when these cultivars grown in the southern provinces, where the onion growing from sets is concentrated, more earliness is expected and accordingly less infection occurs.

Because of the poor keeping quality and the high percent of internal doubling and splitting of the bulbs of the Beth Alpha cultivar, efforts are going on to improve these characters by crossing with the good keeper cultivar, shandaweel.1., and selecting for bulbs with one growing center.

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ABSTRACT

Solarization of soil is under evaluation in Victoria for control of root disease including white rot of onions. A major factor under study is the relationship between distribution of the pathogen in soil after treatment and the effect on disease.

In vitro tests, simulating field conditions, showed that sclerotia of S. cepivorum in soil had a specific heat sensitivity ranging between 40°C and 45°C.

The effect of solarization on distribution of S. cepivorum was studied in field soils artificially inoculated with high levels of sclerotia. Solarization for four weeks in mid summer in S. Victoria, eliminated to 5 cm and reduced to 15 cm the number of viable sclerotia recovered from soil. In S. Victoria the treatment also reduced the number of plant deaths by 21% and increased yields from 18.3 g to 94.1 g.

After treating naturally infested soil at Colac (W. Victoria), solarization reduced the number of viable sclerotia in the 0-20 cm layer of soil by 53%, reduced plant deaths by 34% and increased yields by 19%.

Solarization had the added advantages of controlling most annual and perennial weeds, involved no chemical residues or phytotoxic effects and was non hazardous to use.

Solarization of soil is under evaluation in Victoria as a treatment for control of root disease. The main objective is to determine whether the treatment has significant commercial application. The research includes considerations of the influence of type of pathogen, soil type and climatic zones on the efficacy of solarization. Sclerotium cepivorum is included in the project, and preliminary studies have shown that solarization has potential for control of white rot.

In vitro tests determined that S. cepivorum had a specific heat sensitivity in soil of between 40°C and 45°C. Experiments showed that it was possible to obtain these temperatures in the surface layers of soil solarized in mid summer, with higher temperatures being recorded in Northern Victoria and on heavier soil types.

In 1981 experiments were established at research stations in Northern and Southern Victoria with soil artificially infested with S. cepivorum (Approx. 1000 sclerotia/kg soil). The results showed that in comparison to controls solarization reduced the number of viable sclerotia recovered from soil to a depth of 15 cm in S. Victoria and 30 cm in N. Victoria.

In further studies an experiment was established on a grey sand at the Vegetable Research Station (S. Victoria) to determine the effect of solarization on white rot of onions using artificially infested soil (300 sclerotia/kg soil). Two sets of PVC cylinders containing inoculated soil were placed vertically into moist cultivated soil with the tops level with the soil surface. Small cylinders (10 cm dia. x 32 cm high) were used to determine the distribution of viable sclerotia after treatment and larger cylinders (30 cm dia. x 32 cm high) to assess disease. Once buried within plots (10 m x 1.5 m), the treated plots were covered with 50 µm polyethylene while control plots remained uncovered. The trial design was a randomized block with treatments replicated eight times. After solarizing for four weeks

in mid summer the plastic was removed and the small cylinders were recovered, sectioned transversely at 5 cm intervals, a thin layer of soil sampled and the sclerotia recovered by wet sieving and the viabilities determined, (Porter and Merriman 1983). The larger cylinders were left in the field and onions cv. Savages Flat White were sown into the soil within the perimeter of the cylinder. Plant deaths were monitored regularly during the growing season and assessed for symptoms of white rot.

In comparison with controls solarization eliminated to 5 cm and reduced to 15 cm the number of viable sclerotia recovered from soil (Table 1). Solarization also reduced the number of plant deaths (Fig. 1), with a corresponding increase in the yield of onions harvested within the cylinders and also an increase in bulb size (Table 2). At harvest 93% of control plants were infected with white rot and 74% of treated plants. The high incidence of disease in treated plots (Fig. 1) suggests that the sclerotia surviving treatment at a depth of 10-15 cm in soil are still capable of causing severe disease on onions. The results also suggest that in areas where higher temperatures are recorded at a greater depth in soil, that is, in areas with higher ambient temperatures and/or heavier soil type; or in areas with lower inoculum levels, then solarization would prove a more effective treatment.

The effect of solarization was also determined at a naturally infested field site at Colac (W. Victoria). Plots (5 m x 1 m) were prepared as a seed-bed, then treated with 50 μ m polyethylene from 19/12/80 to 16/1/81. Soil in control plots remained untreated. The trial design was a randomized block with treatments replicated 6 times. Soils were sampled before and after treatment to determine the number of viable sclerotia. After treatment the ground was kept weed free until five rows of onion seed cv. Pukekohe were sown on 16th July, 1981. Plant counts were recorded at emergence and plant deaths were monitored at monthly intervals during the growing season and assessed for symptoms of white rot. At maturity the incidence of white rot and total weight of healthy bulbs/plot were determined.

The results showed that in comparison with controls solarization had reduced the number of sclerotia in the 0-20 cm layer of soil by 53%. The treatment also reduced plant deaths by 34%, with a corresponding increase in yield of 19%. (Table 3).

Solarization had the added advantages of controlling most annual and perennial weeds, involved no chemical residues or phytotoxic effects and was non-hazardous to use.

References

Porter I. J. and Merriman P. R. (1983). Effects of solarization of soil on nematode and fungal pathogens at two sites in Victoria.

Table 1. Effect of solarization on distribution and number of viable sclerotia of S. cepivorum in a grey sand at Frankston in 1982.

Treatment	Depth (cm)	Soil temperature (Absolute maximum)	% viability of sclerotia (100 g) ⁻¹ soil
NIL	3-5	41.7	97.9
	13-15	31.7	100
	23-25	27.8	100
SOLARIZED	3-5	53.9	0**
	13-15	40.6	72.1**
	23-25	34.4	97.6

Significantly different from controls *P = 0.05 **P = 0.01

Table 2. Effect of solarization on the yield of onions grown in soil inoculated with S. cepivorum at Frankston in 1982.

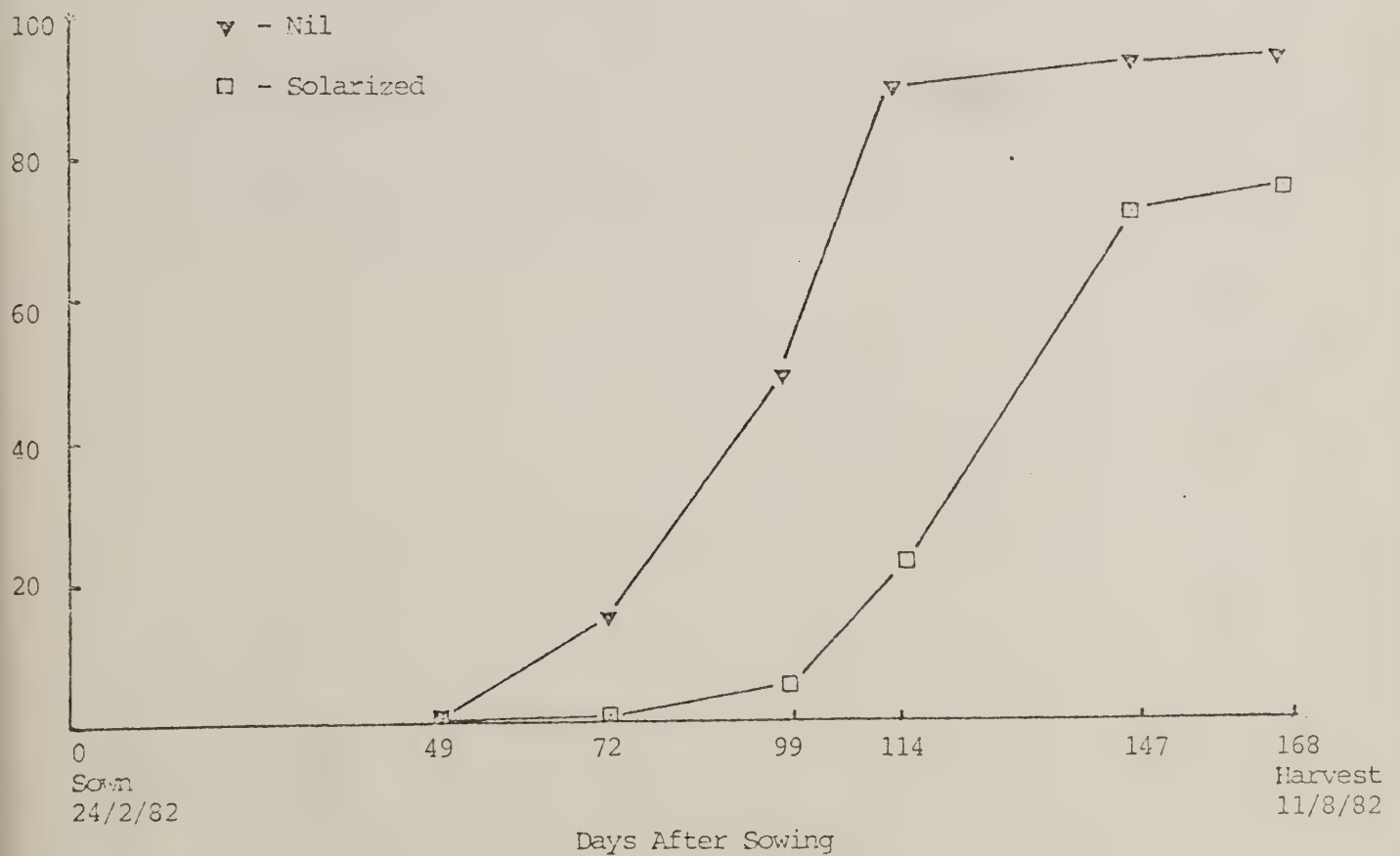
Treatment	Yield/plot (g)	Wt/bulb (g)
NIL	18.3	11.1
SOLARIZED	94.1	18.0

Table 3. Effect of solarization on the numbers of sclerotia, incidence of white rot, and yield of dry bulb onions.

Treatment	Overall % of		
	No. of sclerotia kg ⁻¹ soil (0.20cm)	plants killed due to white rot	Yield of healthy bulbs (t.ha ⁻¹)
NIL	87	53	5.2
SOLARIZED	41**	35*	6.2

Significantly different from control *P = 0.05; **P = 0.01

Fig 1: Effect of solarization of soil on the death
of onions caused by S. cepivorum (Frankston, S. Vic.)



SOIL SOLARIZATION - A POTENTIAL METHOD FOR CONTROLLING WHITE ROT AND OTHER
SOILBORNE DISEASES IN ONIONS.

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|--|---|-----|-----|-----|
| (1) | (1) | (1) | (2) | (2) |
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Solar heating of soils (solarization) is a new method for controlling soilborne pests in hot climate countries. This is accomplished by covering (tarping, mulching) the soil with transparent polyethylene during the hot season, thereby increasing soil temperatures and killing pests. This method, first developed in Israel, is or was examined in additional 13 countries. Its effectiveness against many pathogens and weeds in a variety of crops was shown. For example, excellent control of root and bulb diseases of onions (e.g. pink root, Rhizoctonia, Fusarium) or garlic (Ditylenchus dipsaci) as well as many weeds was shown. The yields were doubled or more. Preliminary experiments for controlling the white rot disease by soil solarization were carried out at Suhag in Upper Egypt. This soil treatment reduced the disease by 50-67%. Addition of Trichoderma reduced the disease by 84-87%. The yields were not increased, apparently due to the light disease infection.

THE EFFECT OF A HOT WATER, FORMALDEHYDE TREATMENT FOR
CONTROL OF STEM AND BULB NEMATODE, ON THE
VIABILITY OF SCLEROTIA OF SCLEROTIUM CEPIVORUM

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INTEGRATED CONTROL SESSION

IN CALIFORNIA, WHERE STEM AND BULB NEMATODE (DITYLENCHUS DIPSACI) IS OFTEN A MAJOR PROBLEM IN GARLIC, A PROCEDURE THAT ERADICATES THE PATHOGEN FROM PLANTING STOCK HAS BEEN DEVELOPED AND IMPLEMENTED. CLOVES ARE IMMersed IN WATER AT 38°C FOR 20 MINUTES. THIS PRE-SOAK IS FOLLOWED BY A SOAKING IN 1% FORMALDEHYDE PLUS DETERGENT AT 49°C FOR AN ADDITIONAL 20 MINUTES.

THE EFFECT OF THE NEMATODE CONTROL TREATMENT ON SCLEROTIA OF SCLEROTIUM CEPIVORUM THAT MAY BE PRESENT ON PLANTING MATERIAL WAS INVESTIGATED. SCLEROTIA ENCLOSED IN SMALL NYLON MESH BAGS WERE TREATED IN THE 1% FORMALDEHYDE WATER AND DETERGENT SOLUTION FOR VARIOUS LENGTHS OF TIME AT 49°C. HEAT ALONE IS DETRIMENTAL TO SCLEROTIA AFTER 20 MINUTES EXPOSURE, BUT THE TIME REQUIRED TO KILL SCLEROTIA WHEN FORMALDEHYDE IS PRESENT DECREASES TO BETWEEN 12.5 AND 15 MINUTES. AS THE TEMPERATURE DECREASES BELOW 47.5°C, THE NUMBER OF SCLEROTIA REMAINING VIABLE EVEN IN THE FORMALDEHYDE, INCREASES.



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